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Analysis of Swiss swine influenza virus genomes

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Abstract

The aim of this work was to complete for the first time a full set of Swiss Swine Influenza Virus (SSIV) genome sequences. A previously published method (Zhou et al., 2009, J. Virol. 83, 10309-10313) for multi-segment amplification of influenza genomes was used in combination with a panel of 49 SSIV isolates collected between 2004 and 2010.
A total of 55 sequences were obtained, covering 15 isolates, 9 representing strains that had previously been propagated in cell culture and 6 derived directly from nasal swabs of pigs with influenza-like disease symptoms.
Analyses of the sequences indicated that the SSIV strains were clearly distinct from the recent pandemic H1N1 strain (Mexico 2009) and were related more closely among each other than to a well-characterized European SIV (Haseluenne 2003). All isolates, of which the neuraminidase (NA) sequence could be determined, were Oseltamivir resistant (H274Y mutation in NA) and featured the R194G mutation in NA, a prerequisite for the consecutive H274Y mutation. Moreover, the predicted membrane protein 2 (M2) aa sequences suggested resistance against the drug Amantadine.
Although the pandemic H1N1 circulated in Switzerland already in 2009, it was observed only in 2011 to entering the Swiss pig population, suggesting that this virus actually represents zoonotic features, though, in the case of Switzerland, not originating from pigs and transmitted to humans but rather contrary, originating in humans and transmitted to pigs.

Introduction

Influenzaviruses

The Influenza A viruses play a prominent role in medicine and veterinary medicine for their ability to infect a broad range of hosts, including a variety of mammals and birds. The influenza viruses belong to the family *orthomyxoviridae*, which is formed by spherical to pleomorphic, enveloped virions with a diameter of 80 to 120 nm, which harbour a nucleocapsid of helical symmetry (Fig. 1). The linear, negative-sense, single-stranded RNA genome is segmented. The family further divides into five genera: Influenza A virus, Influenza B virus, Influenza C virus, thogotovirus, and isavirus. The envelope of Influenza A virions is densely covered with two types of glycoproteins, hemagglutinin (HA or H) and neuraminidase (NA or N). These glycoproteins are traditionally used to define Influenza A virus subtypes, which comprise at present 16 subtypes according to HA and 9 according to NA. In theory, these subtypes can freely reassort due to the segmented nature of the viral genome. More details about the Influenza A virus genome and its encoded proteins are presented further down (Tab. 1).

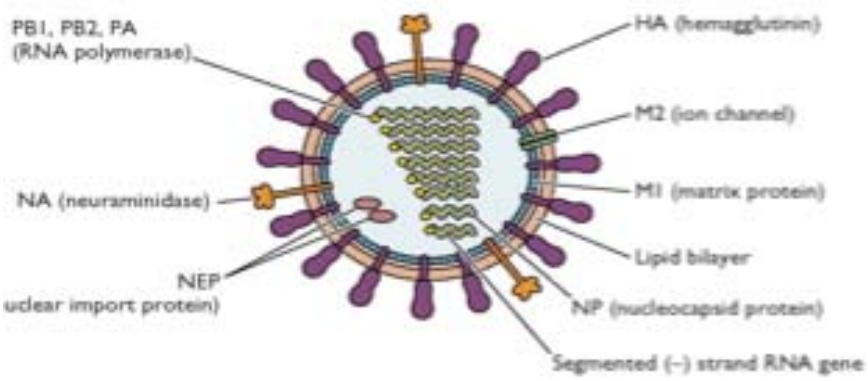

Fig. 1. Structure and components of the Influenza A virus particle. (Source of the picture: Flint SJ, Enquist LW, Racaniello VR, Skalka AM. Principles of Virology: Volume I: Molecular Biology. 3 ed: ASM Press; 2008 (1).)

Influenza A Virus Genome

The influenza virus genome is segmented and consists of a distinct number of single stranded RNA molecules of negative polarity. The influenza A virus genome comprises 8 segments, most of which encode for just one single protein. At least three segments encode for more than one protein. A summary of segment sizes and encoded proteins is provided in Tab. 1

Tab. 1. Genome segments and encoded proteins (2)

No	Size	ORFs: nt	Proteins: aa, MW	Functions
1	2341	PB2: 2277	PB2: 759, 96 KDa	Binds cap of host's pre-mRNA; role in initiation of transcription
2	2341	PB1: 2271	PB1: 757, 87 KDa	Catalyzes attachment of nucleotides; binds vRNA and cRNA for initiation of transcription and replication; generates capped primers for mRNA synthesis
		PB1-F2[a]: 261	PB1-F2[a]: 87, 14 KDa	Sensitizes cells in response to apoptotic stimuli
3	2233	PA: 2148	PA: 716, 85.5 KDa	Component of RNA-polymerase; proteolytic activity; essential for replication and transcription
4	1778	HA: 1650	HA: 550, 76 KDa	Receptor-binding; pH-induced fusion during entry; essential for particle forming and budding; target of neutralizing antibodies
5	1565	NP: 1494	NP: 498, 55 KDa	Encapsidates viral RNA; relevant for RNA transcription, replication and packaging; contains three NLS; responsible for nuclear import of RNA.
6	1413	NA: 1362	NA: 454, 70 KDa	Cleaves sialic acid to release new virus during exit; facilitates entry
7	1027	M1: 756	M1: 252, 28 KDa	Interacts with RNPs and NEP to bring viral components to the plasma membrane; essential for formation of virus particles and budding
		M2: 27+264 [b]	M2: 97, 15 KDa	Ion channel activity; important for uncoating
8	890	NS1: 690	NS1: 230, 25 KDa	Multifunctional: inhibits host mRNA translation; regulates viral pre-mRNA splicing and translation; interferon-antagonist
		NS2: 30+363[b]	NEP: 131, 14 KDa	Nuclear export of viral RNPs
		NSP: 648[c]	NSP: 216, 25 KDa	Transmembrane protein

[a] not present in all influenza A viruses

[b] Emerging by splicing

[c] ORF in the positive sense orientation; typically not present or truncated in avian isolates (43)

Influenza A Viral Proteins

The influenza A virus genome encodes for at least 10 essential proteins, whose functions are briefly summarized in Tab. 1. Some relevant additional information is provided below.

The viral envelope holds two viral glycoproteins, the hemagglutinin (HA) and the neuraminidase (NA) as well as a minor component M2.

- At approximately 500 copies per virion, HA constitutes the major surface glycoprotein. The molecule can be divided into three domains, an ectodomain of approximately 512 aa, a 27 aa transmembrane domain, and a 10 aa cytoplasmic domain. HA homotrimerization is necessary for its receptor-binding activity.

Furthermore, HA undergoes proteolytic cleavage in order to trigger the virus's final infectivity. To this end, the HA-precursor (HA0) is cleaved into HA1 (319 to 326 aa) and HA2 (221 to 222 aa), which activates a conformational change that is a prerequisite for the fusion between the viral envelope and cellular membranes. Between one to six residues are lost during this process. The cleavage itself may take place at different stages of the virus's assembly and maturation process, i.e. still within the virus-producing cell; at the surface of the producing cell, in the environment, and, rarely, within the acidified endosomal vesicle during entry of the newly infected cell.

- NA is the second most abundant spike protein in the viral envelope. The molecule consists of a head domain that is enzymatically active, and a stalk region that is inserted in the viral envelope with a transmembrane region and a short cytoplasmic tail. NA forms homotetramers. Early during infection, the enzymatic activity (acylneuraminyl hydrolase) facilitates transport of the virion through the respiratory tract's mucin layers. Late, NA cleaves sialic acid by which HA remains attached to the cellular receptor, to complete exit and release of the newly formed virus particles. NA co-determines the antigenic type of the influenza viruses and is a major target for antiviral drugs.

- M2 is an integral membrane protein that emerges through splicing of mRNA transcribed from segment 7. Although abundantly produced in the infected cell, only 20 to 60 molecules are incorporated into the virion. M2 forms disulfide-linked homotetramers with a short extracellular domain, a 19 residue transmembrane domain, and a 54 residue cytoplasmic tail. The protein has a low pH-induced ion channel activity that conducts protons to the interior of the particle, which is essential for uncoating. Therefore, M2 constitutes a target for antiviral drugs.

The matrix protein M1, also encoded by segment 7, underlies the viral envelope, providing rigidity. It interacts with the cytoplasmic tails of HA, NA, and M2 on one side and with the ribonucleocapsid (RNP) on the other side. M1 has a nuclear localization signal (NLS) and plays a role in exporting RNP from the nucleus to the site of virus assembly.
The nucleocapsid protein NP is the major structural protein that interacts with the RNA segments to form the RNP and has two NLSs, which enable it to interact with several members of the importin protein family. NP is essential for the synthesis of full-length viral RNA and is, moreover, a type-specific antigen and a target for cross-reactive cytotoxic T-lymphocytes.
The heteromeric RNA transcriptase complex (PB) consists of three individual proteins: PB2, PB1, and PA. Each 30 to 60 copies of these proteins are located in the interior of the virion. Upon infection, they are translocated to the cellular nuclei, where they direct viral gene expression and genome replication.
NS1 and NS2 (NS2 also known as NEP) are both encoded in segment 8 but NS1 is translated from an mRNA transcript, which is colinear with segment 8, whereas NS2 emerges from a spliced mRNA. The two proteins share the first 9 amino-terminal aa but their functions are quite different. NS1 has not been detected in virions but has multiple functions in the infected cell, including RNA-binding and inhibiting splicing and nuclear export of poly(A) containing cellular mRNAs. Ultimately, these activities can prevent the host's interferon response. NS2 exists at an average of 130 to 200 molecules per virion, where it associates with M1 and NP. In the infected cell, the protein localizes to the

nucleus, where it executes an important role for the export of RNPs out of the nucleus. This latter activity has led to its designation as nuclear export protein (NEP).
The functions of two additional influenza virus proteins, PB1-F2 and NSP, are not yet fully understood nor are they known to be essential for influenza virus replication.

Influenza A virus replication

An overview of the viral replication cycle is provided in **Fig. 2**. (Source of the picture: Flint SJ, Enquist LW, Racaniello VR, Skalka AM. Principles of Virology: Volume I: Molecular Biology. 3 ed: ASM Press; 2008 (1))

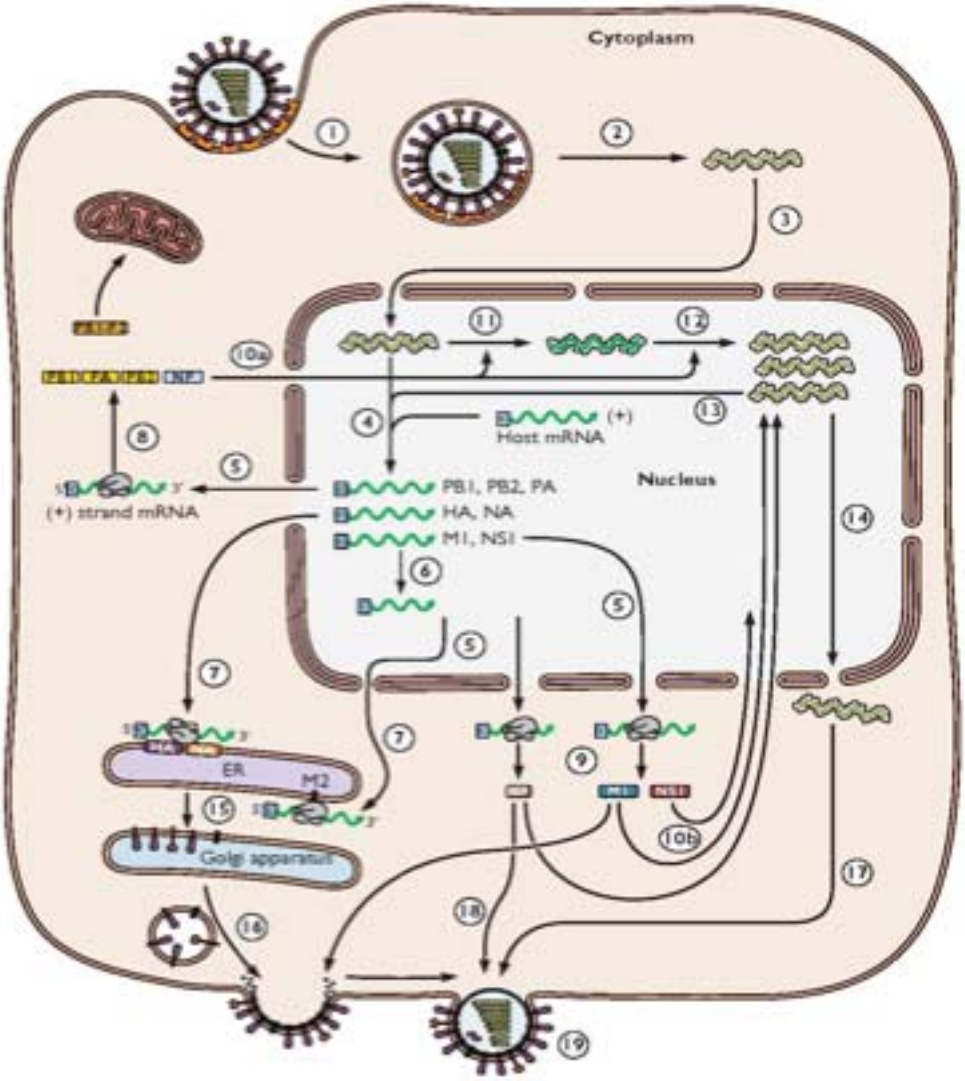

Fig. 2. **Influenza A virus replication cycle**. (1) The virion binds to a sialic acid-containing cellular receptor and enters the cell via receptor-mediated endocytosis. (2) Upon acidification of the vesicle, the viral membrane fuses with the membrane of the vesicle, releasing the eight viral nucleocapsids into the cytoplasm (for simplicity, only one is shown). (3) The viral nucleocapsids containing (-) strand genomic RNA, multiple copies of the NP protein, and the P proteins are transported into the nucleus. (4) The (-) strand RNAs are copied by virion RNA polymerase into viral mRNA, using the capped 5´ ends of host pre-mRNAs (or mRNAs) as primers to initiate synthesis. (5) The mRNAs are transported to the cytoplasm, (6) following splicing in the case of the mRNAs encoding NEP (NS2) and M2. (7) The mRNAs specifying the viral membrane proteins (HA, NA and M2) are translated by ribosomes bound to the endoplasmic reticulum (ER). (8 and 9)

These proteins enter the host cell's secretory pathway, where HA and NA are glycosylated. All other mRNAs are translated by ribosomes in the cytoplasm. (10a) The PA, PB1, PB2 and NP proteins are imported into the nucleus, (11) where they participate in the synthesis of full-length (+) strand RNAs and then (12) of (-) strand genomicRNAs, both of which are synthesized in the form of nucleocapsids. (13) Some of the newly synthesized (-) strand RNAs enter the pathway for mRNA synthesis. (10b) The M1 protein and the NS1 protein are transported into the nucleus. (14) Binding of the M1 protein to newly synthesized (-) strand RNAs shuts down viral mRNA synthesis and, in conjunction with the NEP protein, induces export of progeny nucleocapsids to the cytoplasm. (15) The HA, NA, and M2 proteins are transported to the cell surface and (16) become incorporated into the plasma membrane. (17) The virion nucleocapsids associated with the M1 protein and the (18) NEP protein are transported to the cell surface and attach to regions of the plasma membrane that contain the HA, NA, M1, and M2 proteins. (19) Assembly of virions is completed at this location by budding from the plasma membrane.(1)

Evolution and Pandemic Outbreaks

The general reservoir of the Influenza viruses is in various avian species. Thus, all influenza viruses originally descend from avian Influenza viruses. All the known HA and NA subtypes have been detected in these animals and evolutionary stasis indicates that the avian influenza virus subtypes have adapted optimally to their hosts. In general, the same viruses are less stable in mammalians than in avians, a fact that can be measured by the mutation rates affecting the individual segments. In the mammalians, the mutation rates affecting the segments encoding for HA or NA are quite high, whereas they are relatively low in the segments encoding NP, M1, and the components of the viral RNA-polymerase.

Influenzavirus genomes are known to evolve either in great leaps through reassortment and recombination or gradually through the accumulation of point mutations. Reassortment is based on the simultaneous infection of the same cell by two different Influenza viruses. Upon replication and packaging, the genomic segments of the two co-infecting viruses can be randomly mixed, i.e. reassorted. Reassortment of the segments encoding for either HA or NA may lead to dramatic antigenic changes of the viral progeny, which is described by the term antigenic shift. A similar or lesser effect can be the result of recombination among the genomes of two different parental Influenza viruses, an effect that is attributed to the copy-choice mechanism. Accumulation of point mutations in the segments encoding for HA or NA may lead to the so-called genetic drift effect. Both antigenic shift and antigenic drift provide means for the virus to survive and evolve in the face of strong immune responses.

All these processes may lead to increased virulence and fitness of the selected viruses. Thus, sometimes they have been hold responsible for leading to epidemics or pandemics. Indeed, antigenic shift is believed to being the basic prerequisite for several Influenza A virus pandemics.

Spanish Influenza

Although pandemics had been recorded earlier, the so called Spanish Influenza of 1918 was the first one of which the agent could be reconstructed. This pandemic appeared in three waves. In spring the outbreak started with a very mild but highly contagious form of disease, showing but a low mortality rate. The first reported cases were traced back to American soldiers in Fort Riley, Kansas, where more than 1000 cases were recorded

within one month. The disease then spread to other army camps and around the world with the mass movement of soldiers in connection with World War I. In August a much more virulent form of the virus emerged, causing an enormous mortality rate in the U.S.A. and Europe. The peak of the pandemic was reached in November, followed by a third wave, which lasted until spring of 1919. The official death toll amounted to 25 million people.
Viral replication was restricted to the respiratory tract, causing pneumonia with secondary bacterial infections and finally respiratory failure leading to death. There was no proof for systemic infection. In contrary to later pandemics, the Spanish influenza affected mainly young adults, mostly sparing children and elderly people.
The causative agent was identified as an H1N1 influenza virus. Upon sequencing of the genome, which was possible only decades later, it became clear that this virus had its origin in aquatic birds but represented a combination of genomes from human and avian virus progenitors. Interestingly, the viral genome did not carry typical indicators of virulence but harbored a mutation changing the codon 190 into aspartic acid (D) in HA, which facilitated binding to human receptors.

Russian Influenza

In 1977 another H1N1 pandemic was recorded, affecting mainly younger people under 25 years of age. The official death toll amounted to approximately 700'000 people. Older people were resistant, probably due to pre-existing immunity. Due to its close relationship to the 1918 virus, it has been speculated that the agent of this pandemic may have escaped from a laboratory.

Asian Influenza

A novel H2N2 virus was the cause of a pandemic in 1957. It is believed that this virus emerged as a reassortant between the circulating human H1N1 and an avian H2N2 virus. Not only HA and NA but also PB1 were shown to be of avian origin. Although the virus did not appear to be very aggressive, it spread rapidly worldwide and took a death toll of approximately one million people, which was attributed to have resulted from a lack of pre-existing immunity.

Hong Kong Influenza

This pandemice H3N2 virus was first recorded in 1968 in China and seemed to represent a novel reassortant with H3 and PB1 segments originating from avians. Thanks to the pre-existing immunity against N2, the death toll did not get above approximately 700'000.

Human Bird Flu

In 1997 a novel, high pathogenic avian H5N1 virus first emerged in Asia. This virus, also known as human "bird flu", spread rapidly worldwide by means of migrating birds. Surprisingly, this virus can be transmitted directly from chicken to humans and other mammals. Fortunately, there was no evidence for human to human transmission. However, the same virus returned in 2002, 2003, and 2006, taking its toll with case fatality rates in humans of up to 60%. Not only humans were affected by the associated disease but also more than 130 species (including chickens, swans, pigeons) and mammals (various felines, ferrets, dogs, pigs, donkeys). Remarkably, this virus was not confined to the respiratory tract but was also recovered from cerebral fluid and stool. The H5N1 virus was found to be resistant against several antiviral drugs (3).

Outbreak of pandemic H1N1 influenza in 2009 in Mexico

In April 2009 the Pan American Health Organisation was informed about a severe outbreak of influenza in a province of Mexico. A novel H1N1 influenza virus was found to be responsible. Apparently, this virus was a reassortant of several viruses circulating in pigs in America and could have been transmitted to humans several months earlier. The

disease spread quickly in Mexico and only a few days later the first cases appeared in USA.

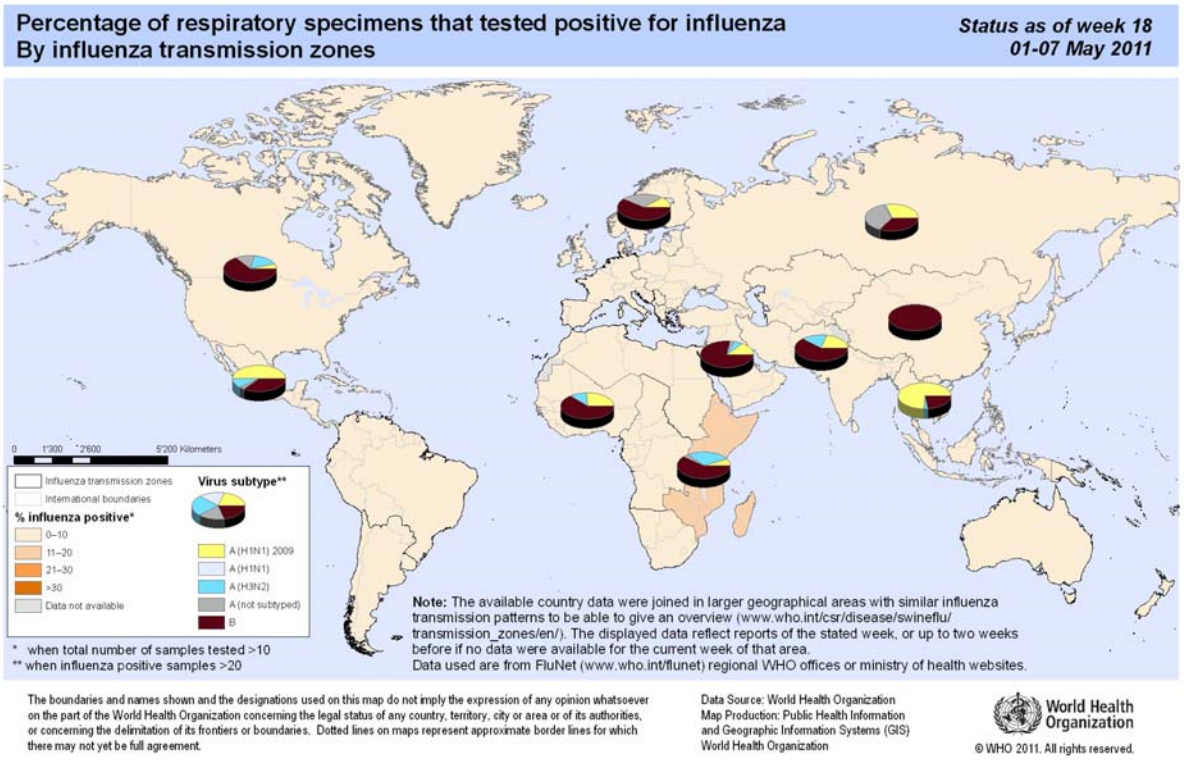

Fig. 3. **Status of influenza in May 2011.** The map shows the percentage of respiratory specimens, which were tested positive for influenza in the different influenza transmission zones, and the distribution of different virus subtypes in the positive tested. (Source of the picture: World Health Organisation Map Production Public Health Information and Geographic Information Systems (GIS), WHO. Update Nr. 134, May 20, 2011)

Fortunately, the death rate remained relatively low with an average global case fatality rate of between 0.01-0.03%. The whole genomic sequence of the new virus was determined very rapidly, thus, making accurate RT-PCR-based diagnostics possible. Until December 2009, 47 millions of Americans were infected. 9'820 patients died from Swine Flu, whereas 213'000 had to visit a hospital. Overall this strain of influenza virus caused (yet) a very mild course of illness with a relatively small number of deaths. From the USA the virus was further spread by travellers to Europe and Asia, causing a classic pandemic. Meanwhile, the novel H1N1 virus, also termed "Swine Flu", has established an endemic state in many countries worldwide (Fig. 3).
Similar to the Spanish influenza, the disease was more risky for the people at age of less than 65 years than to the older ones. But contrary to 1918, children were at high risk. In the end of December 2009 number of cases started to decrease and by Spring 2010 the pandemic was considered completed.

Pathogenesis and disease in avians, pigs, humans and horses

The original reservoir of influenza virus is in aquatic birds, which can be infected by all subtypes. However, influenza A viruses are known to infect as well a broad range of other birds and mammals, including for example humans, swine, horses, dogs, cats,

whales, and seals. Some subtypes have successfully established their independent circulation and evolution among those latter animal species. Originally, the influenza viruses have evolved to replicate in the epithelial cells of aquatic bird intestines and to be shed in faeces. During enteric replication, the virus makes predominantly use of the so-called avian receptor, a terminal sialic acid (SA) linked to galactose by an alpha 2,3 linkage. However, depending on the availability and density of the receptor, influenza virus may also replicate in the respiratory tract. For successful entry and productive replication, the receptor-binding protein, hemagglutinin (HA), needs to be cleaved by host proteases into HA1 and HA2. The sequence of the cleavage site as well as the availability of suitable proteases are both important for the virulence of a specific influenza virus strain in birds.

So-called low pathogenic (LP) avian influenza virus strains possess a single Arg at the cleavage site. Such HA molecules are not cleaved by intracellular proteases but depend on the presence of extracellular trypsin-like proteases, which limits the spread of those viruses within the organism. However, the virulence of LPAI can be aggravated by certain bacteria, which secrete suitable proteases or induce local accumulation of host thrombin or plasmin, both of which can cleave HA of LPAI.

In contrast, highly pathogenic (HPAI) viruses feature several basic amino acids at the HA cleavage site, which will be cleaved by ubiquitous intracellular subtilisin-like proteases, such as furin. These viruses can infect a broad range of host cells, causing fatal systemic infection in poultry. Such viruses also make use of a broader receptor range, including SA linked to galactose by an alpha 2,6 linkage, which is abundant in the respiratory tract of a broad range of mammals. The ability of using the 2,6 receptor (also termed the human receptor) dramatically increases the risk of transmission to humans. Furthermore, the systemic spread within the infected organism also opens new routes for transmission for these viruses. Feeding off carcasses from HPAI-infected birds is a well-known source for the infection of carnivores, which then also may succumb to influenza (4, 5, 6 ,7, 8, 9).

Compared to the avian species, mammals are not considered to function equally well as influenza A virus reservoir. As a rule of thumb, influenza viruses from one species do not easily replicate in other species. However, some subtypes of avian influenza A have crossed the species barrier for good and established pandemics as well as subtype-specific reservoirs in their new hosts. Aside of humans, the most important examples comprise pigs (H1N1 and H3N2), horses (H7N7 and H3N8), and seals (H7N7) (10, 11, 12).

In mammals, the influenza A viruses target rather the respiratory tract than the enteric tract of their hosts and the infection is mainly transmitted by the respiratory route (13, 14). Systemic infections are very rare. Pigs are considered potentially important for the development of pandemic influenza virus strains because of the following reasons: (1) their upper respiratory tract contains an abundance of both avian and human influenza virus receptors, which makes them susceptible for all sorts of influenza A viruses. (2) Indeed, pigs are considered equally susceptible for avian and human influenza virus strains, which may gradually lead to the evolution of viruses that make more efficient use of the human receptor. (3) After co-infection of avian and human influenza viruses, reassortant viruses may emerge, which may be highly pathogenic for humans and carry the potential to cause pandemic outbreaks.

Presently, at least 3 subtypes of influenza A viruses are known to circulate among pigs. A classical H1N1 swine influenza A virus is a leading cause for a transient respiratory illness in pigs (15). More recently, an H3N2 virus with human-like HA and NA genes and an avian-like H1N1 have established endemic states among pigs (10, 16). Occasionally,

human influenza A viruses are transmitted to pigs (and vice versa) and reassortants originating from those as well as from the various endemic swine influenza viruses are known to emerge (17). Influenza in pigs is highly contagious and associated with high fever (up to 42°C), anorexia, apathy, and respiratory symptoms such as laboured breathing and coughing, rhinitis, sneezing. (13). Occasionally, conjunctivitis and reproductive problems may be observed. However, subclinical infections may also occur. Usually, the diseased pigs recover from the illness within 6 days. (13).
Humans are not only susceptible to influenza A viruses but also to influenza viruses B and C (18, 19, 20). However, only influenza A will be discussed in the present context. The predominant subtypes circulating among humans include combinations of H1, H2, H3, N1, and N2. However, H5N1, an entirely avian virus, was first observed in 1997 to successfully transgress from birds to humans and causing fatal disease (21). Direct transmissions to humans of avian H7N7 and H9N2 have also been reported, although without such a severe outcome as with H5N1 (22, 23).
The clinical signs in adults vary from asymptomatic to severe, fatal pneumonia but are, in general, similar to those observed in pigs. Respiratory signs include rhinorrhea, sneezing, and dry coughing. Systemic symptoms, due to the innate immune responses, include headache and high fever. Inflammation of the conjunctivae may also be seen. The duration of disease may take up to two weeks.
In children, the clinical signs may be more severe and may include higher body temperature elevation, otitis media, and myositis. In severe cases an interstitial pneumonia may develop. Complete regeneration can take up to one month. Furthermore, a higher incidence of gastrointestinal and severe central nervous symptoms is noted among infected children. The detailed pathogenesis of the CNS symptoms, which may range from confusion to coma, in children is not known. However, they are believed to base on non specific metabolic reactions due to pneumonia, rather than systemic infection (24, 25, 26).
Just one main subtype of influenza A does presently circulate among horses, namely H3N8 (influenza A equi 2) (11). H7N7, also known as influenza A equi 1, is presently considered to be extinct (27). Influenza virus spreads rapidly among horses in affected premises and is easily transmitted to neighbouring stables. The clinical symptoms (i.e. pyrexia, dry coughing, rhinitis) are similar to those observed in pigs. The disease is mostly self-limiting unless complications due to secondary bacterial infections occur. Cases of heart muscle inflammation have been reported in the context of H3N8 infections (28). Frequently, vaccinated horses undergo merely subclinical infections. However, vaccination does not limit the spread of equine influenza. Under the pressure of vaccination, equine influenza evolves by antigenic drift but, interestingly, is not known to be transmitted to other animal species.

Diagnostics

The diagnostic methodology applied for the detection of influenza virus varies certainly with the purpose of the diagnostic procedure. While rapid tests are preferred in the clinical context, the detection of novel influenza viruses as well as diagnosis in the epidemiological context have a preference for tests that will provide as much information as possible about the virus of interest. For a very long time, virus isolation has been considered the gold standard of influenza virus diagnostics (29, 30, 31). Newly emerging test procedures were, therefore, routinely assessed against virus isolation. Positive test results that had yielded negative virus isolation were, therefore, habitually considered "false positive". However, due to their sophistication and rapid performance,

molecular diagnostic approaches are increasingly able to compete with the original gold standard.

Virus isolation. Classically, influenza viruses have been isolated by inoculating embryonated chicken eggs. For this purpose, fertilized eggs from SPF hens were incubated until day 11. Subsequently, a small volume of virus was inoculated into the allantoic cavity of the egg. After two days of further incubation, the virus was harvested for its specified use. Alternatively, the virus can be isolated by inoculating cell cultures. For example, chicken embryo fibroblasts, primary epithelial cells of human adenoid, primary monkey kidney cells or cell lines such as Vero cells, MRC-5, Madin-Darby kidney cells (MDCK) are commonly used for isolating influenza viruses. (31, 32, 33, 34).

Detection of influenza virus proteins. Hemagglutinin-inhibition (HI) as well as complement fixation (CF) tests have been traditionally used to identify influenza viruses either directly in clinical samples or else after amplification in cell cultures (35). CF was considered as broadly applicable for screening because it targeted the conserved nucleoprotein (NP). However, the test was limited in its results because it may be able to discriminate between Influenza virus types A, B, and C, whereas the influenza virus subtypes cannot be addressed with this method. Alternatively, the HI test is suitable for subtype testing because the targeted antigen is the hemagglutinin (HA). Indeed, for its high reliability the HI test remains useful up until today. More recently, antigen detection has shifted to more sensitive methods, such as immunofluorescence (IF) and ELISA applications. The targeted antigens include either NP as well as the highly conserved matrix protein (M1) for rapid influenza virus typing or HA and neuraminidase (NA) for subtyping (29, 36). More recently, several biosensor-based technologies have been developed to rapidly and sensitively detect influenza virus proteins in complex samples (31). However, none of these latter approaches has yet made it to the market.

Detection and characterization of influenza virus RNA. Nowadays, various forms of reverse transcription (RT) followed by polymerase chain reaction (PCR) are routinely used for influenza virus diagnostics. The amplification products can be sequenced in order to further characterize positive samples. Of course, the usability and complexity of the obtained sequences depends largely on the genomic loci targeted and the lengths of the amplification products. From very early on, it was clear that those methods equalled or surpassed all other methods by their sensitivity, versatility, and ability to shorten the time needed for an appropriate diagnosis (31, 36, 30). More recently, these methods were refined to multiplexing and use of microarray applications (37, 38, 39). These latter methods are designed to simultaneously identify on an analytical level and with a high sensitivity and specificity the presence of various types and subtypes of the influenza virus.

Full genomic sequence determination. It is very clear from the above considerations that the informative value of any diagnostic procedure increases with the amount of genomic sequence that is generated throughout. Indeed, at least two related approaches have been published to amplify and sequence entire influenza virus genomes, even without the need to first amplify the viruses in cell culture (40, 41). Both methods take advantage of the highly conserved genomic packaging sequences that are present close to the ends of each influenza virus genomic segment. These conserved sequences make it possible to amplify, clone, and sequence the entire genome of any influenza A virus. Thanks to omitting the need for a step involving cell culture (needing 4-10 days), the procedure can be completed in a very short time and provides a maximal amount of information about the virus of interest within 24 to 48 hours.

Sensitivity and specificity. Systematic assessments of the various diagnostic procedures are difficult to achieve because uncertainties are inherently involved starting from the quality of sampling, transport, initial handling in the laboratory, and so on, up to the suitability of cell cultures and reagents in relation to each diagnostic event. However, according to the literature (31, 36), the sensitivity of PCR-based assays ranges around 5 $TCID_{50}$, whereas isolation by cell culture requires 10^4 $TCID_{50}$, and ELISA-based assays need even higher amounts of virus to produce a positive result.

Surveillance of Influenza in Switzerland

In Switzerland, influenza is monitored separately for humans (Laboratory of Virology, University of Geneva Hospitals), birds (National Reference Center for Poultry and Rabbit Diseases, University of Zurich), and pigs (Reference Center for Swine Influenza, Institute of Virology, University of Zurich). However, the three participating laboratories promote close contacts and routinely exchange technical information.

In 2009, 141 medical practitioners participated in an official monitoring program for human influenza. They reported the number of consultations with patients suffering from fever of more than 38°C, myalgia, coughing, and rhinorrhea. Moreover, 91 of these practitioners submitted nasal swabs from their patients to the Reference Centre for Influenza in Geneva. In total, 4893 samples were tested. 1470 of them were positive for influenza virus. 1442 were positive for influenza A (H1N1), 22 were diagnosed as seasonal influenza and 3 of them were influenza B.

On 26 of April 2010, the WHO declared the outbreak of influenza pandemic, which became later known as the "pandemic H1N1 swine flu". The first case in Switzerland was detected on 28 April 2010 in a human returning from Mexico. Thereafter, the virus circulated among the Swiss population between May and August of the same year with a peak at the beginning of July. As a consequence of the pandemic, the influence virus detection methods were adapted accordingly. Until February of 2011, a total of 13441 samples were tested positive for the pandemic influenza virus strain.

Influenza in pigs has been monitored systematically since 2004. The program is being supported by both the Federal Office of Health (BAG) and the Federal Veterinary Office (BVet). Collection of the samples was predominantly contributed through practitioners of the Swiss Swine Health Service (SGD). Samples were to be taken from farms with pigs showing influenza-like symptoms, such as fever, coughing, and nasal discharge. Annually, a collection of 150 to 250 samples came together, covering 35 to 77 farms from all over Switzerland. The frequency of influenza-positive samples collected from 2008 to 2010 amounted to around 50% from approximately 66% of the farms tested. While it has not been possible to precisely subtype each case, all positive cases represented influenza A viruses. Approximately 50% of the positively identified viruses were successfully propagated in cell cultures. All viruses that had been successfully subtyped were H1N1, while neither H3 nor N2 were ever detected (Annual reports 2008-2010). The samples from 2009 and 2010 were, moreover, specifically assayed for the presence of the pandemic H1N1 virus but all with negative results. Thus, there was no evidence that this pandemic virus circulated among the Swiss pigs prior to 2011. In contrast, the limited number of sequences achieved through these PCR methods, indicated that the influenza viruses circulating in the Swiss pig population were most closely related to common European H1N1 strains. However, a full set of genomic sequence from any Swiss porcine influenza virus has not been reported prior to this work.

Purpose of the present work

The circulation and identity of potentially zoonotic viruses among farm animals is of public interest. One possibility of monitoring such viruses is by serology, the other is by characterization of their genomes. However, serological methods also require that the corresponding agent is known and well characterized.

In the present work, we set out to characterize influenza A viruses circulating among Swiss pigs (Swiss Swine influenza viruses; SSIV). Since isolation of those viruses by inoculation of cell cultures was frequently unsuccessful, we decided to establish a single reaction, multi-segment RT-PCR (41), which had been designed to amplify, clone, and sequence the entire genome of any influenza A virus, without the need to propagate it first in cell culture. Using this method, we wanted then to determine the sequences of as many SSIVs as possible in order to characterize them and to compare the sequences to those of other influenza A viruses, in particular those of European Swine influenza viruses and also the 2009 pandemic H1N1 virus, which had been claimed to have originated in pigs.

Working hypothesis. MS-RT-PCR will facilitate cloning and sequencing of SSIV genomes and their comparison to European Swine influenza viruses as well as to the 2009 pandemic H1N1 virus.

The expectation was that the results of this work would provide for the first time a full set of SSIV genomic sequence and allow its positioning among other well characterized influenza viruses. In the future, these sequences might also be used for generating antigens either for establishing serological tests or for producing vaccines.

Materials and Methods

Viruses. A total of 49 swine influenza virus strains, collected between 2004 and 2010 in the context of our swine influenza surveillance program, were used in this study. Details are listed in Table 2. The laboratory strain A/PR/8/34 (kindly provided by J. Pavlovic, Med. Virology, University of Zürich) was used as a control. For RNA extraction, we either used supernatant from nasal swabs or from infected cell cultures.

Cell culture. For virus propagation, we used the MDCK cell line (ATCC), grown with Iscove's Modified Dulbecco's Medium (Fluka/Sigma Chemie AG, Buchs, Switzerland), containing 7% foetal calf serum (Amimed, BioConcept, Allschwil, Switzerland) and antibiotic, antimycotic solution 100x (Fluka/Sigma).

Virus propagation. MDCK cell monolayers were inoculated with supernatant from nasal swabs (first cell culture inoculation) or with infected cell culture supernatant (further cell culture passages). The inocula were diluted 1:2 with serum-free Iscove's Modified Dulbecco's Medium containing 1.25 µg/ml trypsin (bovine pancreas, Fluka/Sigma). After inoculation, the cells were incubated for 90 minutes at 37°C with Serum-free medium containing 1.25 µg/ml trypsin. Then, the cells were further incubated at 37°C for 72h. After two cycles of freezing / thawing, the supernatants were clarified from cellular debris by centrifuging at 3000xg for 10 minutes. Finally, 1 ml aliquots were stored at -80°C until further use.

RNA extraction: RNA was extracted from 140 µl swab or cell culture supernatant using the QIAamp Viral RNA Mini Kit (QIAGEN AG, Hombrechtikon, Switzerland) according to the manual provided by the manufacturer.

MSRT-PCR: The method has been described by Zhou et al. (2009) and was adapted by testing different conditions as described below (Optimization). The final protocol, using the Superscript III One-Step RT-PCR with Platinum Taq Polymerase Kit (Invitrogen/Life-Technologies, Basel) was performed in a total volume of 40µl, consisting of 20µl of "2x-Reaction-Mix ", 1µl Forward Primer MBT uni-12 (3'-ACGCGTGATCAGCAAAAGCAGG-5', 0.8 µM), 1µl Reverse Primer MBT uni-13 (3'-ACGCGTGATCAGTAGAAACAAGG-5', 0.8µM), 2µl Platinum Taq Mix, 16µl RNA. Run conditions: 42°C for 60min, 94°C for 2min, followed by 36 cycles 94°C for 30s, 45°C for 30s, 68°C for 3min.

Optimization of the MSRT-PCR conditions. Nineteen different primer- concentrations from 0.05 to 2µM were tested. Furthermore, the influence of the magnesium-concentration was analyzed by using 1.6M, 1.7M, 1.8M, 1.9M or 2M solutions. The effect of DMSO (5%) and betaine (0.5M), was measured either alone or in combination. To find the optimal annealing temperature, the PCR was run using a twelve-step gradient from 45°C to 60°C. The final elongation time was increased from three to five minutes.

Reverse Transcription and PCR. For cDNA production, the Reverse Transcription System (Promega AG, Dübendorf, Switzerland) was used with the Uni12 primer 5'-AGCAAAAGCAGG-3') according to Hoffmann et al. (2001). The cDNAs were purified using the QIAquick PCR Purification Kit (QIAGEN) and eluted in 30 µl elution buffer. Both procedures were done according to the instructions of the manufacturers. The amplification of segments 1, 2 and 3 (PB2, PB1, PA) was conducted as described by Hoffmann et al. (2001), using a primer concentration of 3 µM, 8 µl of cDNA template, and the Expand High Fidelity PCR System (Roche Diagnostics, Rotkreuz, Switzerland).

Gel electrophoresis and DNA extraction. The PCR products were analyzed on 2% agarose gels and DNA of PCR products, cut out from the agarose gels, was extracted using the QIAquick Gel Extraction Kit (QIAGEN). The Quick-Load 1 kb DNA ladder (New England BioLabs GmbH, Brüningstrasse 50, Geb. B852, 65926 Frankfurt am Main) was used as MW marker.

Cloning. Extracted PCR products were cloned into the pCR 2.1 TOPO Vector, using the Topo TA Cloning Kit (Invitrogen) according to the provided instruction manual. DNA, extracted from positive clones by a standard mini prep procedure, was tested by EcoRI restriction enzyme analysis to identify clones with desired insert sizes.

Sequence analysis. The cloned PCR products were sequenced in both orientations by Microsynth AG (Balgach, Switzerland), using standard M13 primers. The resulting sequences were assembled with ContigExpress (Vector NTI). For completing the entire sequence of the single segments, follow-up primers were designed based on the sequencing results. Sequences were compared with influenza virus sequences deposited in the Genebank by BLAST analyses (NCBI database). Occasionally, the sequences could not be resolved properly. In these cases, we manually corrected individual nucleotides according to the sequence of its closest neighbor, making use of either other SSIVs or the Haseluenne strain. These manual corrections are listed in Appendix 9.

Alignments. Alignments were done with Align X (Vector NTI). The Guide Tree calculations were based on a sequence distance method and utilized the Neighbor Joining (NJ) algorithm of (42) Comparison with foreign strains was done with BLAST (National Center for Biotechnology Information).

Results

Experimental design.

In order to get for the first time a complete set of at least one Swiss Swine influenza virus (SSIV) isolate, a total of 49 historic as well as newly incoming samples were subjected to multi-segment amplification. For cloning the segments coding for the polymerase, a segment-specific RT-PCR was taken to achieve enough PCR-product to clone the bands. Amplified fragments were cloned and subjected to nucleotide sequence analysis. Verified sequences were aligned among each other as well as in comparison with at least one other European SIV and in comparison with the recent pandemic H1N1 virus. Similarly, the translated amino acid sequences were also aligned and analyzed with respect to eminent protein signatures.

Optimization of multi-segment amplification (MSRT-PCR).

After an initial screening (data not shown), it turned out that the MSRT-PCR as described by Zou et al.
needed some optimization to work properly in our hands. Therefore, sufficient template was produced from strain SIV18/10 ZP2, which performed well in cell culture. Using this template, the following parameters were individually modified: primer concentration, magnesium concentration, elongation time, addition of betaine and DMSO, and annealing temperature. Fig. 4 shows that the optimal primer concentration ranged between 0.2 and 2 µM.

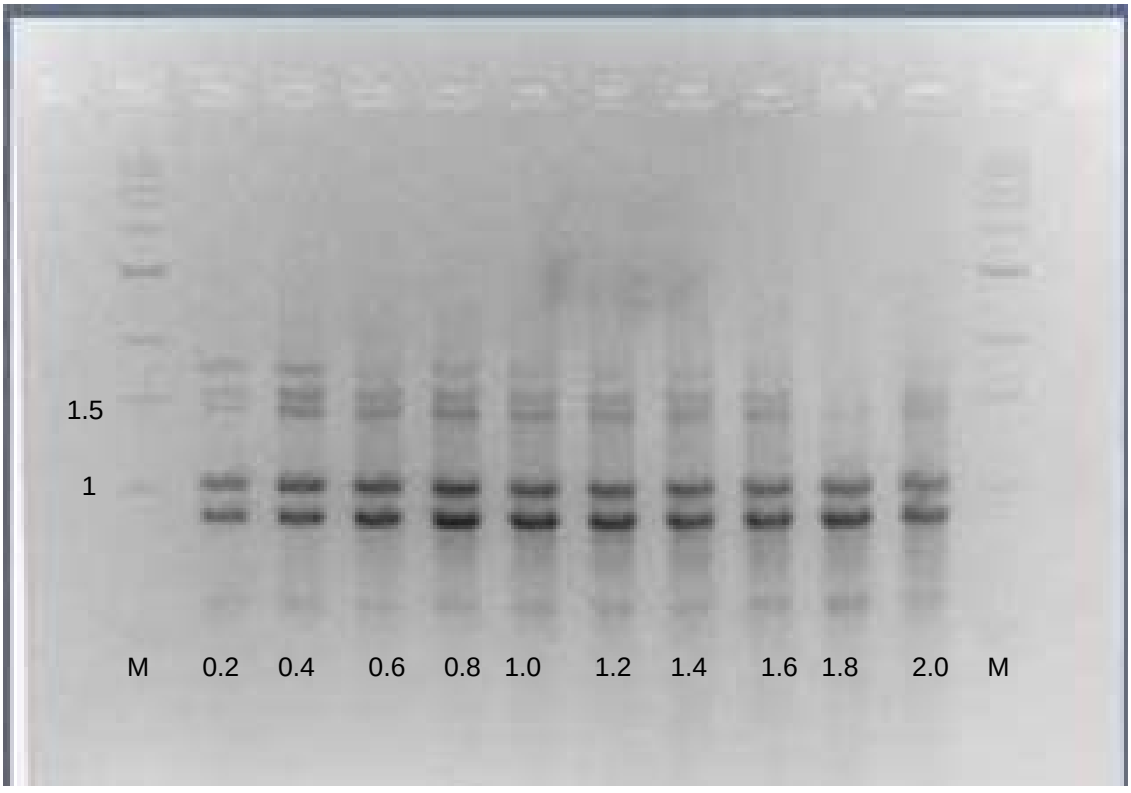

Fig. 4. Agarose gel electrophoresis of amplification products after MSRT-PCR of SIV 18/10 ZP2. Increasing primer concentrations (µM range) were used as indicated on the bottom of each lane. The Quick-Load 1 kb DNA ladder was used as MW marker (first and last lane). The 1.5 and 1 kbp bands of the marker are indicated on the left.

The highest band intensity was obtained using 1.6 M magnesium, whereas addition of betaine and/or DMSO had a negative effect on the band intensities (data not shown). Increasing the annealing temperature did not fundamentally affect the number of amplified bands, although some variation in band intensity was observed (Fig. 5).

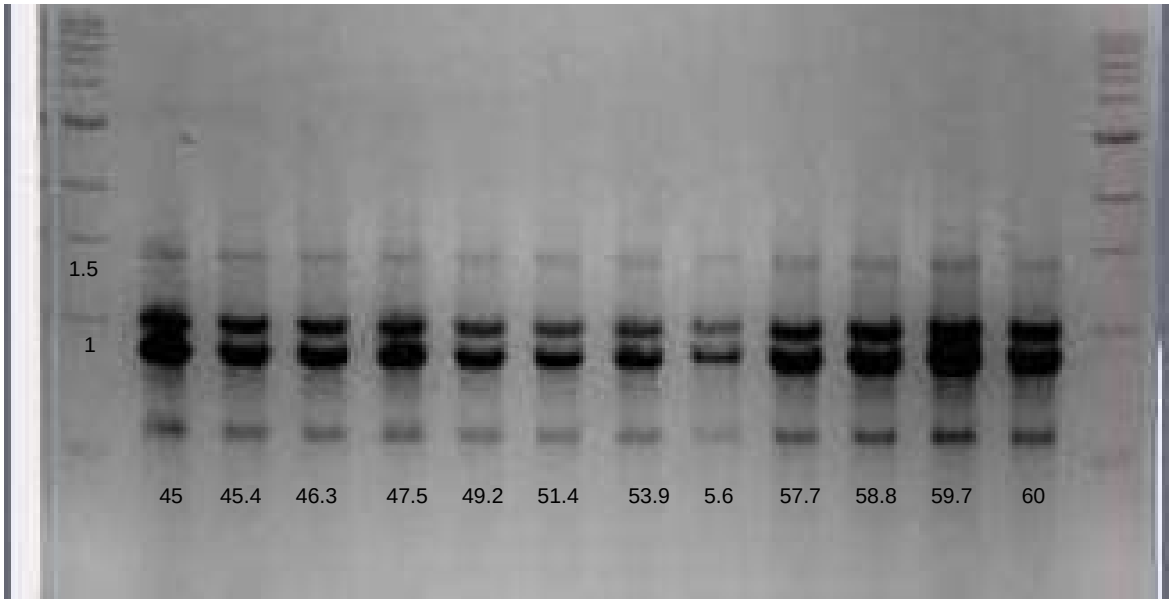

Fig. 5. Agarose gel electrophoresis of amplification products after MSRT-PCR of SIV 18/10 ZP2. A gradient in annealing temperature (°C range) was used as indicated on the bottom of each lane. The Quick-Load 1 kb DNA ladder was used as MW marker (first and last lane). The 1.5 and 1 kbp bands of the marker are indicated on the left.

However, increasing the elongation time from three to five minutes had a negative effect on the intensity of the slowly migrating bands (Fig. 6).

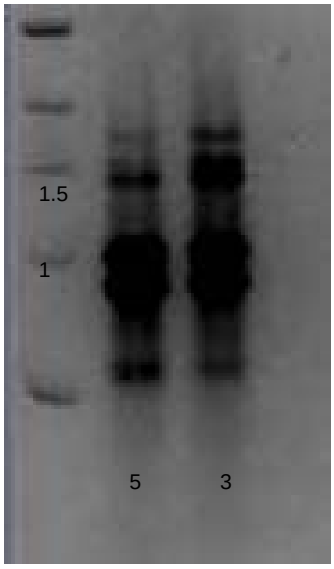

Fig. 6. Agarose gel electrophoresis of amplification products after MSRT-PCR of SIV 18/10 ZP2. Two different annealing temperatures of three and five minutes were tested, as indicated on the bottom of each lane. The Quick-Load 1 kb DNA ladder was used as MW marker (first lane). The 1.5 and 1 kbp bands of the marker are indicated.

In summary, a primer concentration of 0.8 µM, Mg concentration of 1.6, elongation time of three minutes and an annealing temperature of 45°C provided the highest number of bands at high intensity.

MSRT-PCR of Swiss SIV isolates.

Based on the above results, all samples as well as the laboratory strain (PR8) were subjected to MSRT-PCR. For this purpose, templates were extracted from 20 swab samples and from 30 cell culture supernatants.

Tab. 2 summarizes the results of these experiments and gives also an overview of the sample's designation and origin. Briefly, no band was amplified from 6 samples, i.e. two swabs and four cell culture supernatants. As a rule, smaller segments were more readily amplifiable than larger segments. Only one segment (segment 7) was amplified from one single sample, whereas all other samples provided amplification of multiple segments, i.e. 12 times two segments, 4 times three segments, 7 times four segments (including with strain PR8), 15 times five segments, and 5 times all eight segments. Thus, an important precondition to sequence at least one full set of SSIV segments had been achieved.

Tab. 2. Overview of strain designation, origin, fate of sample, and analyses done.

Strain [a]	**Canton** [b]	**Sample** [c]	**Amplified Segments** [d]	**Cloned and sequenced Segments**
PR8	n.a.[e]	c	8,7,6,5,4	-
SIV 54/04	BE	c	8,7,6,5,4	8,7
SIV 42/05	BE	c	8,7,6,5,4	-
SIV 25/06	ZH	c	8,7,6,5,4	7
SIV 01/09	BE	s	7,5	-
SIV 04/09	BE	s	-	-
SIV 14/09	AI	c	8,7	-
SIV 60/09	BE	s	7,5	-
SIV 63/09	SG	s	7,5	-
SIV 67/09	ZH	c	-	-
SIV 73/09	BE	c	8,7	-
SIV 88/09	NE	c	8,7,6,5,4	8

SIV 94/09	AG	s	8,7,5	-
SIV 98/09	TG	s	8,7,5	-
SIV 126/09	SG	c	8,7,6,5,4	-
SIV 132/09	SO	c	8,7,6,5,4, (3,2,1)	8,7,6,5,4
SIV 146/09	BE	c	8,7	-
SIV 151/09	LU	c	8,7	-
SIV 157/09	BE	c	8,7,6,5,4	8,7
SIV 170/09	LU	c	-	-
SIV 180/09	SO	c	-	-
SIV 186/09	TG	c	8,7,5	-
SIV 194/09	BE	c	8,7,6,5	-
SIV 199/09	AG	c	8,7,6,5,4	8,6,5
SIV 202/09	BE	c	8,7,6,5	-
SIV 206/09	LU	c	8,7,6,5,4, (3,2,1)	8,7,6,5,4
SIV 212/09	SO	c	8,7,6,5,4	-
SIV 216/09	LU	c	8,7,6,5,4	-
SIV 226/09	TG	c	8,7,6,5,4	7
SIV 237/09	TG	c	8,7,6,5,4	8,7
SIV 245/09	ZH	s	-	-
SIV 246/09, 247/09 (same farm)	AG	s	8,7,6,5,4 (3,2,1) [f]	8,7,6,5,3,2
SIV 249/09	VD	s	7,6	-
SIV 262/09	BE	c	8,7,6,5	-
SIV 265/09	SG	c	8,7	-
SIV 268/09	AG	c	8,7	-
SIV 270/09	SG	s	8,7,6,5,4	8,7,6,4
SIV 274/09	BE	c	8,7,6	-
SIV 281/09	AG	c	-	-
SIV 292/09	ZH	s	7,6	-
SIV 294/09	ZH	s	8,7,6,5 (3,2,1)	8,7,6,5,3,2,1
SIV 03/10	SH	s	8,7,6,5,4	-
SIV 15/10	BE	s	7,4	-
SIV 18/10	LU	s	8,7,6,5,4,(3,2,1)	8,7,6,5,4,3,2,1
SIV 21/10	SG	s	7,4,(3,2,1)	-
SIV 33/10	ZH	s	7,4,(3,2,1)	-
SIV 38/10	AG	s	7	-
SIV 42/10	AG	s	8,7,6,5,4	-
SIV 45/10	AG	s	8,7,6,5,4	8,7,6,5,4
SIV 54/10	FR	s	8,7,6,5,4	8,7,6

[a] Strain designation
[b] Origin of sample
[c] Fate of sample: S (nucleic acid extraction directly from nasal swab); C (sample amplified in cell culture)
[d] Segment number that was successfully amplified by MSRT-PCR
[e] not applicable
[f] Segments 1, 2, 3 were visible as a single band upon agarose gel electrophoresis

Cloning and sequence analysis of the amplified segments.

The segments were cloned with TOPO TA Cloning after extracting the bands from the gel. We received 55 sequences, including 13 times each of segment seven and eight, 9 times segment 6, segment 5 was cloned in 7 isolates and segment 4 in 5. The polymerase bands were amplified with the PCR described by Hoffmann (40), what was done with three isolates. We were able to clone three times PBA (segment 3) and PB1 (segment 2). Only two sequences were obtained for PB2 (segment 1). Altogether we succeeded in completing the first full genomic set of a Swiss SIV, namely SIV 18/10. Furthermore, we completed the sequences of segments 4-8 from three isolates. Different combinations of segment 1 to 7 sequences were obtained from the remaining isolates.

Segment 8

Complete nucleotide sequences from segment 8 were obtained from 13 different SSIV isolates. All of them were most closely related to influenza A H1N1. The aligned

nucleotide sequences are shown in Appendix 1a. Although the nucleotide identity among segment 8 of the Swiss isolates was very high (97 to 100%), they seemed to branching into two groups. The more conservative strains originated from the Central and Eastern part of Switzerland (LU, ZH, TG, SG), with only one exception (157/BE). The Western strains (AG, SO, BE, FR, NE) seemed to be more variable and were also on the same branch of the guide tree as SIV Haseluenne 2003 and the pandemic H1N1 influenza virus strain (Mexico 2009).
However, both groups were clearly distinct from Haseluenne 2003 with which they shared an identity level of 92 to 93%. Mexico 2009 was even more distant, showing an 80 to 82% identity with the SSIV on the nucleotide level.

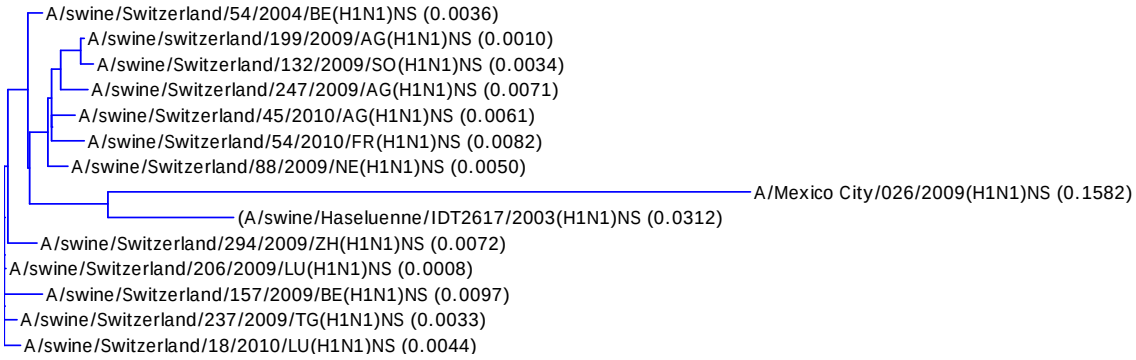

Fig. 7: Guide tree of the alignments of segment 8. The complete segment 8 nucleotide sequences of 13 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 8 is indicated by its products (NS proteins). The calculated distance values are indicated in parenthesis.

The alignment of the predicted NS1 aa sequences (Appendix 1b) revealed that strain Haseluenne 2003 featured a predicted protein of 230 aa. In contrast, all of the SSIV sequences coded for a C-terminally truncated version of the same protein with a length of 217 aa. Similarly, Mexico 2009 featured an ORF encoding for 219 aa protein, which was also truncated relative to Haseluenne 2003. Moreover, both Haseluenne and the pandemic H1N1 distinguished themselves in position 36 (L36) from all of the SSIVs (I36). Yet, Haseluenne differed at position 39 (E/D) relative to pandemic H1N1 and all SSIVs. Finally, a difference was also noted at position 60, where Mexico 2009 encoded for a valin (V60), whereas the SSIVs and also Haseluenne 2003 coded for an alanine (A60). Interestingly, V60 in NS1 is considered as a signature for an influenza virus that is transmitted from human to human (hu-hu), whereas A60 is typical for avian to avian (av-av) transmission. (43).
Overall, the SSIVs branched into two groups, both clearly differing from either Haseluenne or pandemic H1N1. The identities of the Swiss isolates among each other was with 94 -100 % very high. With identities of 84-87% SSIVs were more similar to Haseluenne 2003 than to Mexico 2009, where identities between 79 and 80% could be observed. Hasenlunne 2003 differed even more to Mexico (2009) with an identity of 76%.

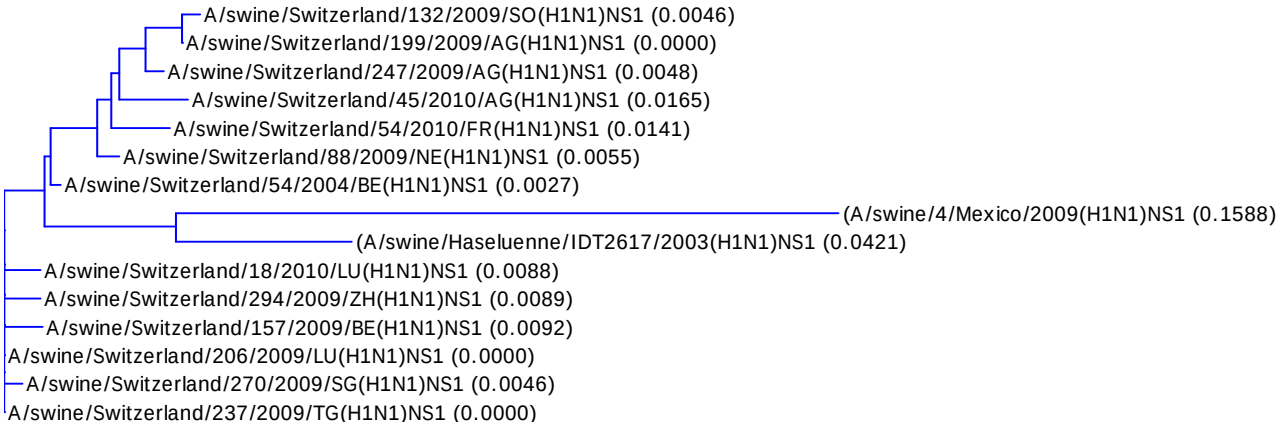

Fig. 8: Guide tree of the alignments of the translated NS1 protein. The predicted NS1 amino acid sequences of 13 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

The alignment of the predicted NS2 aa sequences (Appendix 1c) revealed high conservation among the SSIVs and only few differences compared to either Haseluenne or pandemic H1N1. The identity level among the Swiss isolates amounted to 98 to 100%, i.e. they differed among each other by one or 2 amino acids. Interestingly, one spot of a hu-hu signature was detected at position 70 (G) of the predicted NS2 aa sequences of SSIVs. The highest variation was seen at positions 22 (G22R) and 23 (S23P). SSIV against Haseluenne shared an identity of 96 to 98%, whereas SSIV against pandemic H1N1 amounted to 87 to 88% identity.

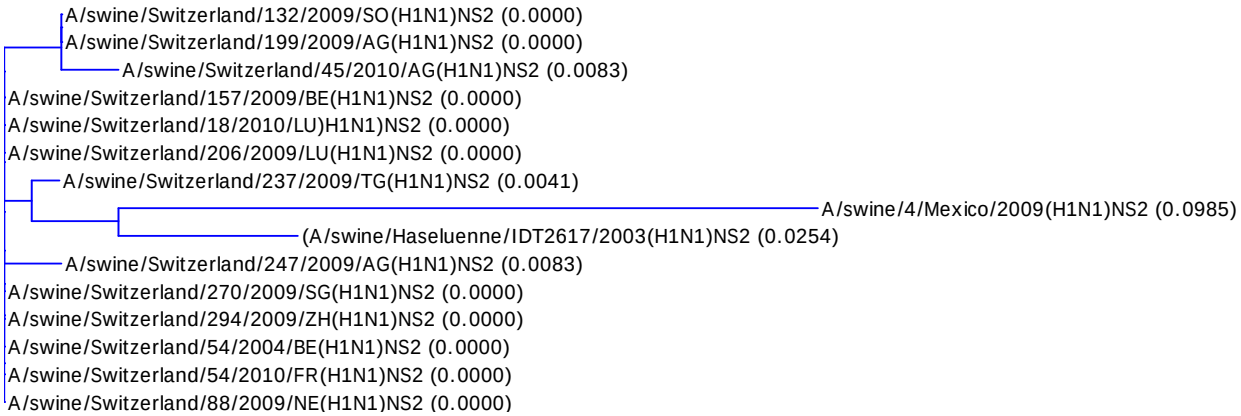

Fig. 9: Guide tree of the alignments of the translated NS2 protein. The predicted NS2 amino acid sequences of 13 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Overall, one may conclude that segment 8 of the SSIVs was highly conserved. Its encoded ORFs and the corresponding aa sequences differed clearly from the European

Haseluenne SIV as well as from pandemic H1N1. The most remarkable feature was a C-terminal truncation of the predicted NS1 aa sequence.
According to Tab. 1, some influenza viruses encode for an additional protein (NSP) (44) translated from the complementary sequence of segment 8. However, all isolates studied here contained early truncations in the predicted NSP sequences. Therefore, this protein is not further addressed in this work.

Segment 7

Segment 7 encodes for two open reading frames, M1 (252 codons) and M2 (97 codons), which slightly overlap. Thirteen nucleotide sequences of segment 7 from SSIV were compared to each other as well as to the sequences of Haseluenne 2003 and the pandemic strain Mexico 2009 (Appendix 2a). All Swiss isolates encoded for the two expected, overlapping ORFs and were very similar compared to each other, with identities of 97 to 100%. However, they branched again into two groups, a more conservative group of Swiss Eastern strains and a more variable group of Swiss Western strains. Haseluenne 2003 was clearly separate from the Swiss strains (identity of 94 to 95%). Mexico 2009 showed only 88 to 89% identity with the Swiss isolates and 91% identity with Haseluenne 2003. Altogether, segment 7 of the SSIV seemed to be well conserved.

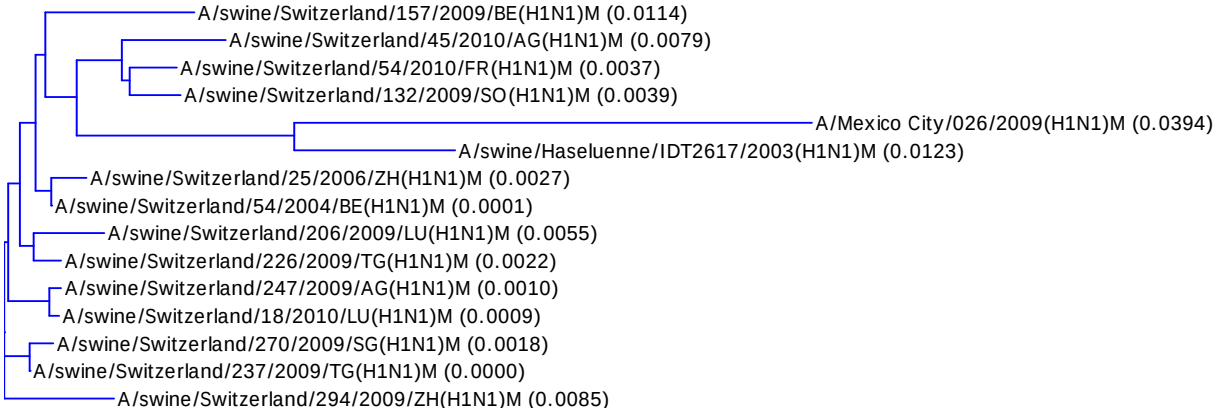

Fig. 10: Guide tree of the alignments of segment 7. The complete segment 7 nucleotide sequences of 13 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 7 is indicated by its products (M proteins). The calculated distance values are indicated in parenthesis.

The predicted aa sequences of M1 were highly conserved (98 to 100% identity) among Haseluenne 2003, Mexico 2009 and, with one exception (SSIV 237/09), all the Swiss isolates (Appendix 2b). Two Swiss isolates, SIV 237/09 and 270/09 (SG) had exchanged the otherwise conserved Leucine at position 103 for Isoleucine. Another potentially relevant mutation was detected in SIV 226/2009 (TG) with the replacement of a polar Threonine by a nonpolar Isoleucine at position 150 (T150I). Finally, with 115V, 121T and 137T, all of the predicted M1 proteins carried the signatures of avian-to-avian (av-av) transmitted Influenza viruses, rather than of human-to-human (hu-hu) transmitted ones (43).

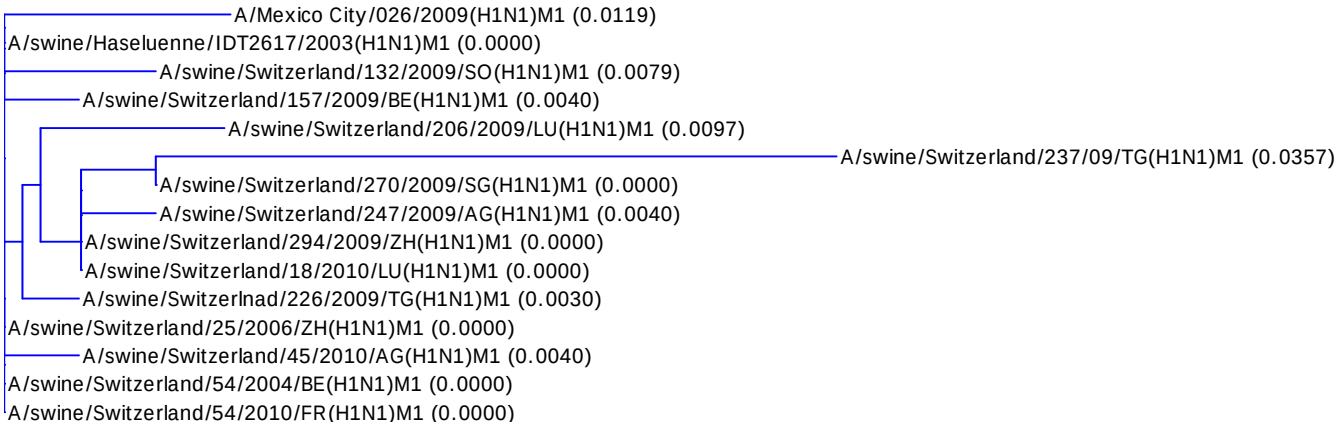

Fig. 11: Guide tree of the alignments of the translated M1 protein. The predicted M1 amino acid sequences of 13 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Only little variation was detected among the predicted aa sequences of the M2-protein. The percentage of identity among the SSIVs ranged between 96 and 100% (Appendix 2c). Indeed, differences of the SSIV aa sequences to the consensus sequence were detected just in 0 to 2 positions. However, Haseluenne 2003 differed by 6aa and Mexico 2009 by 11aa positions. However, all of the strains analyzed here coded for an Asparagine (N) at position 31. Most of the signatures for av-av transmission (11T, 20S, 54R, 57A, 78Q and 86V) remained conserved in the SSIV strains. Only strain 270/2009 (SG) and Mexico 2009 carried a hu-hu signature at position 14 (G14E). Mexico 2009 differed further from the other isolates by coding for a Valine at position 27 and Phenylalanine at position 55. The L55F mutation turned the av-av signature into a hu-hu signature. (43)

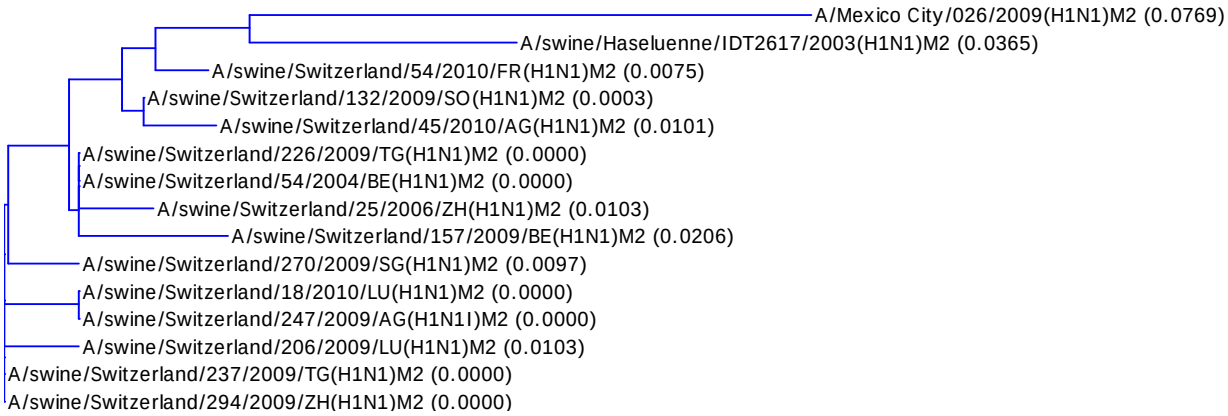

Fig. 12: Guide tree of the alignments of the translated M2 protein. The predicted M2 amino acid sequences of 13 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Segment 6

Segment 6 encodes for the neuraminadase protein (N or NA). Nine nucleotide sequences of segment 6 in SSIVs were compared to each other as well as to the sequences of the Haseluenne strain (2003) and the pandemic strain Mexico (2009) (Appendix 3a). The overall identity of the nucleotide sequences amounted to 82.7%, with a similarity of 99.9%. The Haseluenne and Mexico strains, sharing among themselves 91.1% identity, were clearly distinct from the Swiss isolates, which, among themselves, reached an identity of 93% and a similarity of 100%. The alignment of the Swiss segment 6 nucleotide sequences revealed 63 dispersed loci, where some of the sequences differed from the consensus. Moreover, two distinct groups were detected among the Swiss strains, one heterogeneous group, which was somewhat closer to Haseluenne and Mexico, and a second group, which was more distant with regard to Haseluenne and Mexico but less heterogeneous, sharing among itself more than 97% identity.

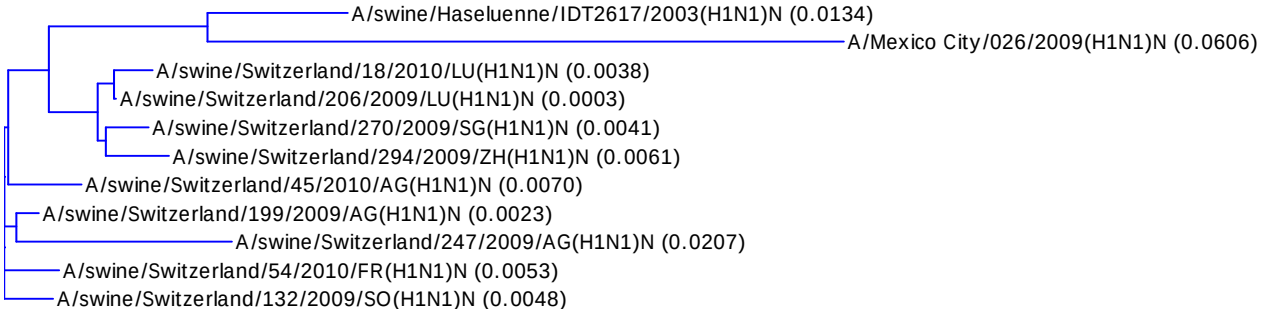

Fig. 13: Guide tree of the alignments of segment 6. The complete segment 6 nucleotide sequences of 9 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 6 is indicated by its product (N protein). The calculated distance values are indicated in parenthesis.

As expected, less variation was observed upon the alignment of the predicted aa sequences (Appendix 3b). However, one group of 4 Swiss isolates (18/2010, 206/2009, 270/2009, 294/2009) featured three sites of discrepancy relative to the consensus sequence, (1) exchange of the polar T at position 20 by a nonpolar A. In strain 270/2009, this was even accentuated through exchange of a second polar amino acid (S) by a nonpolar one (G) at position 21. (2) All four of those isolates also featured an aa exchange at position 83. (3) The consensus isoleucine at position 321 was replaced by valine in three of the four isolates from the diverging group. Interestingly, Haseluenne (2003) grouped in this instance with the diverging isolates, whereas Mexico (2009) featured an isoleucine at this position.

Importantly, all of the isolates analyzed in this set of comparison, including Haseluenne and Mexico, featured the functionally important mutations R194G and H274Y mutation (45).

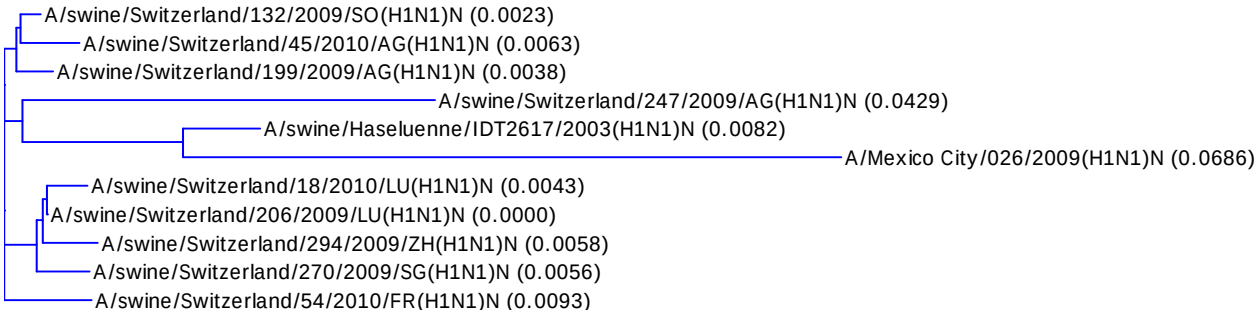

Fig. 14: Guide tree of the alignments of the translated Neuraminidase. The predicted NA amino acid sequences of 9 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. In this case "N" in the figure stands for Neuramindase protein. The calculated distance values are indicated in parenthesis.

Segment 5

Segment 5 encodes for the multifunctional influenzavirus nucleoprotein (NP), whose primary purpose is to encapsidate the viral genome (46). We compared the segment 5 nucleotide sequences from seven Swiss isolates against those of Haseluenne (2003) and Mexico (2009) (Appendix 4a). The consensus level was very high, ranging from 99 to 100%. However, the identity in the alignment of all those strains reached only 77%. Mexico 2009 was clearly more distinct from the SSIV than the Haseluenne 2003 isolate. Indeed, 214 differences inside the NP ORF and 8 differences outside of the ORF were detected between Mexico 2009 and the SSIV. In contrast, the difference between Haseluenne 2003 and the SSIV amounted to 27 positions inside and 3 positions outside of the ORF. All those differences were distributed randomly along the sequence. However, eight loci seemed to be important for the formation of the guide tree.

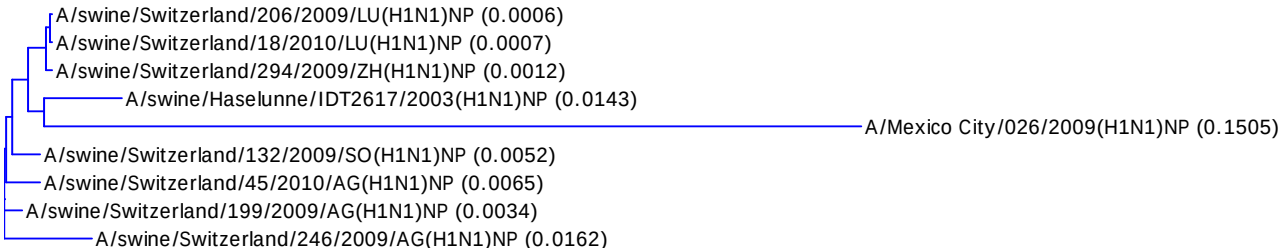

Fig. 15: Guide tree of the alignments of segment 5. The complete segment 5 nucleotide sequences of 7 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 5 is indicated by its product (NP protein). The calculated distance values are indicated in parenthesis.

A similar branching was observed upon alignment of the aa sequences, with an overall 100% similarity and 88% identity positions (Appendix 4b). Individual differences were scattered all along the predicted molecule. Among the SSIVs, all of twelve predicted sites were indicative for av-av transmission. In contrast, both Haseluenne 2003 and Mexico 2009 showed a glycine at position 33 instead of valine, which led to the loss of a

predicted av-av site. Moreover, Mexico 2009 showed R100V and Q357K, which changed the original av-av sites into a hu-hu sites.

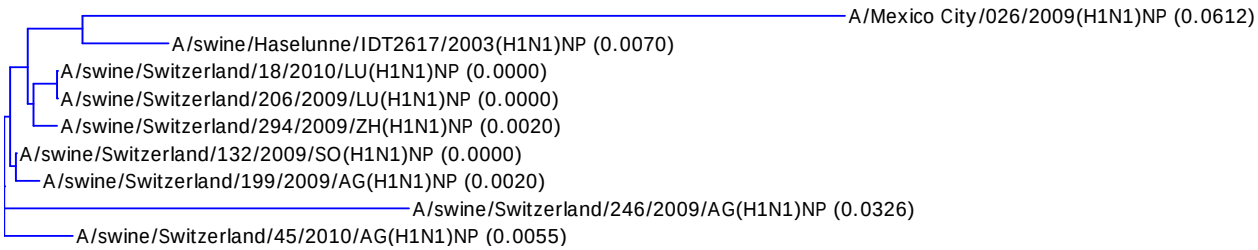

Fig. 16: Guide tree of the alignments of the translated Nucleoprotein. The predicted NP amino acid sequences of 7 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Segment 4

Segment 4 encodes for the influenzavirus hemagglutinin (HA). Importantly, all of the Swiss isolates belonged to the H1 subgroup. On the nucleotide sequence level, an overall consensus of almost 100% was observed with a percentage of 89.5% identity (Appendix 5a). Upon aligning with Mexico 2009 and Haseluenne 2003, the consensus level dropped to 99% and the percentage of identity to 62.5%. This observation suggested that the Swiss isolates were closely related among each other, more different from European H1N1 isolates, and clearly distinct from the recent pandemic H1N1. Differences compared to the consensus sequence were scattered all along the sequences. Interestingly, SSIV 45/2010 showed a slightly longer HA-ORF than all the other isolates.

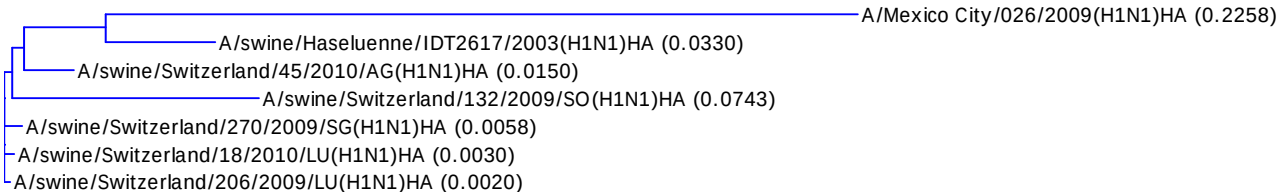

Fig. 17: Guide tree of the alignments of segment 4. The complete segment 4 nucleotide sequences of 5 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 4 is indicated by its product (HA protein). The calculated distance values are indicated in parenthesis.

A similar picture was obvious on the aa sequence level (Appendix 5b). Mexico 2009 diverged in 119 (94 in the HA1 fragment) positions from the consensus, whereas Haseluenne 2003 differed only in 18 positions (15 in HA1). The Swiss isolates differed even less among each other. However, the putative cleavage site between HA1 and HA2, the signal sequence, and the transmembrane region were essentially conserved among the different isolates.
In this regard, only Mexico 2009 showed a few differences compared to the other isolates. SSIV 45/2010 encoded for a lysine (K) at position 39, exactly as Haseluenne

2003 and Mexico 2009. In contrast, the other SSIVs encoded for an arginine (R) at position 39. At position 123, Mexico 2009 and Haseluenne 2003 as well as SSIVs 45/2010 and 270/2009 encoded for a serine (S), whereas SSIVs 206/2009 and 18/2010 encoded for a glycine (G) at the same position. Interestingly, all the strains compared in this study encoded for a Valine (V) at position 190.

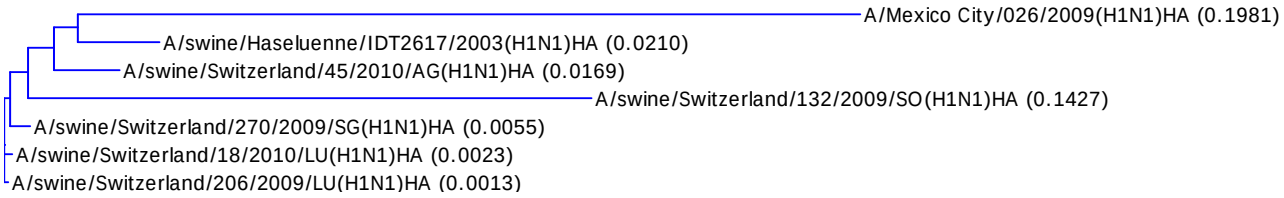

Fig. 18: Guide tree of the alignments of the translated Haemagglutinin. The predicted HA amino acid sequences of 5 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Segment 3

Segment 3 encodes for PA, one of the three viral polymerase subunits. Comparison of segment 3 of three different Swiss isolates against Haseluenne (2003) and Mexico (2009) revealed that the SSIVs were very similar among each other with an identity of 97.2% and a consensus of 100% on the nucleotide level (Appendix 6a). Haseluenne (2003) still shared 94% identity with the SSIVs, whereas Mexico (2009) was further distant with 85% identity shared with the other isolates. Mexico (2009) differed in 300 positions and Haseluenne (2003) in 122 positions from the Swiss consensus sequence. The differences were scattered randomly over the sequence, yet most of them were located within the PA ORF.

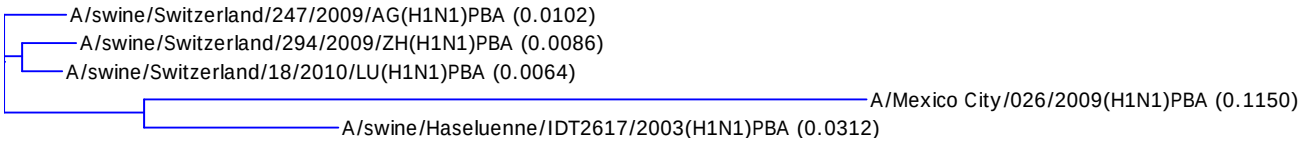

Fig. 19: Guide tree of the alignments of segment 3. The complete segment 3 nucleotide sequences of 3 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 3 is indicated by its product (PBA protein). The calculated distance values are indicated in parenthesis.

Similarly, the predicted aa sequences of SSIV PA were highly conserved with an overall identity of 90.6% and consensus of 99.9% (Appendix 6b). Among 17 positions, which had been mapped to indicate either av-av or hu-hu transmission, the SSIVs showed 11 that were indicative for av-av transmission. The remaining positions were not indicative for either hu-hu or av-av.

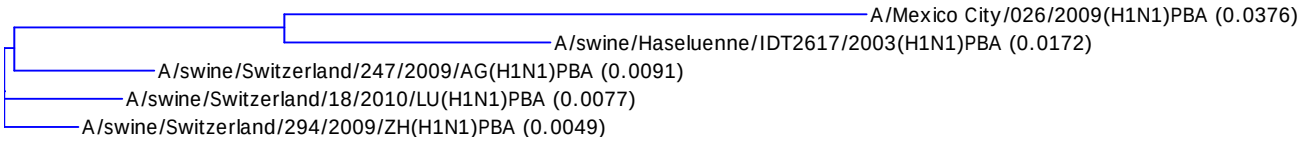

Fig. 20: Guide tree of the alignments of the translated PBA protein. The predicted PBA amino acid sequences of 3 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Segment 2

This segment is bicistronic with a length of approximately 2.2kb and encodes for the viral polymerase subunit PB1 and for the short proapoptotic peptide PB1-F2 (47). Nucleotide sequences of segment 2 of three Swiss isolates as well as of Mexico (2009) and Haseluenne (2003) were compared (Appendix 7a). With 96-99% identity, SSIVs were building one closely related branch. Haseluenne (2003) with 89-91% identity was more distant, while Mexico (2009) was furthest off, sharing 78-81% identity with the Swiss isolates and 85% identity with Haseluenne (2003).
Overall, the segment was quite conserved, showing among the nucleotide sequences of the Swiss isolates only 61 differences against the consensus, all located inside the PB1 ORF but otherwise randomly distributed along the sequence. Haseluenne (2003) showed 96 different nucleotides compared to the SSIVs. One of the differences was located outside of the PB1 ORF. Mexico (2009) showed 334 nucleotides differing from the consensus, two of them outside the ORF.

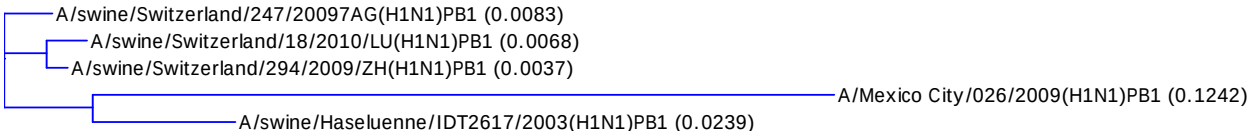

Fig. 21: Guide tree of the alignments of segment 2. The complete segment 2 nucleotide sequences of 3 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 2 is indicated by its product (PB1 protein). The calculated distance values are indicated in parenthesis.

On the aa level of the PB1 ORF, a consensus of 100% was observed with an overall identity of 91.6% (Appendix 7b). The two SSIVs showed a level of 99% identity between each other, 98% identity with Haseluenne (2003), and 95% with Mexico (2009). SSIV 247 (2009/AG) differed at two locations from the consensus sequence within the N-terminal PA-binding domain (aa1 through 87). Furthermore, four differences against the consensus were noticed among all of the SSIVs in the C-terminal PB2-binding domain. At position 336 of PB1, the European strains encoded for a valine, indicating av-av transmission, while Mexico (2009) encoded isoleucine, which is indicative for hu-hu transmission.
A most interesting difference was observed from within the PB1-F2 ORF, which was maintained in two SSIVs as well as Haseluenne (2003) but early terminated in Mexico

(2009). Interestingly, the PB1-F2 ORF was truncated to 79aa in one of the SSIVs (247/2009/AG), whereas the other SSIVs as well as Haseluenne (2003) encoded for a 90 aa peptide (Appendix 7c).

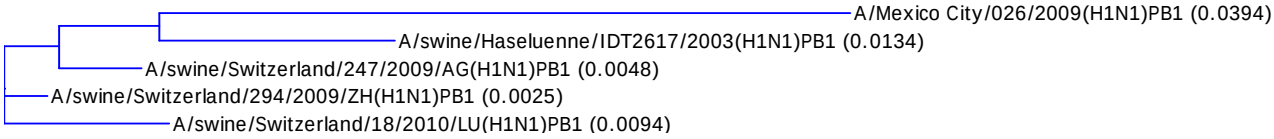

Fig. 22: Guide tree of the alignments of the translated PB1 protein. The predicted PB1 amino acid sequences of 3 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Segment 1

This segment encodes for the viral polymerase subunit PB2. We had two segment 1 sequences of Swiss isolates available for comparison with Haseluenne (2003) and Mexico (2009). On the nucleotide sequence level, the consensus amounted to 100%, whereas the identity was at 80%. Without Mexico (2009), the identity level increased to 96%, whereas it amounted to 98.8% between the two SSIVs (Appendix 8a). A total of 375 differences were detected between the four sequences but only one mapped outside of the PB2 ORF. 26 differences were noted among the SSIVs and 70 in the comparison of the SSIVs to Haseluenne (2003).

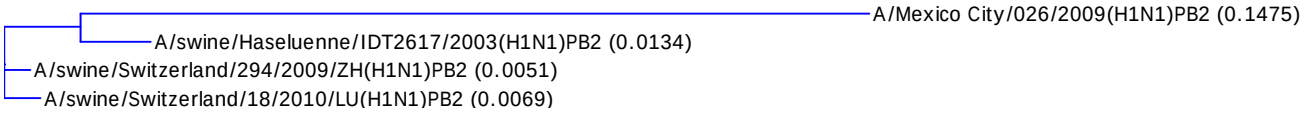

Fig. 23: Guide tree of the alignments of segment 1. The complete segment 1 nucleotide sequences of 2 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name. For shortening, segment 1 is indicated by its product (PB2 protein). The calculated distance values are indicated in parenthesis.

The overall aa consensus between the four isolates amounted to 100%, with 99.3% identity (Appendix 8b). Without Mexico (2009), the identity level increased to 97.2%, while it amounted to 98.9% (7 different aa) in between of the two SSIVs. In six of the seven loci encoding a variation, one of the Swiss isolates differed from the consensus. However, the I649V variation from within the RNA-binding domain was shared between Mexico (2009) and SSIV 249 (2009, ZH). Variation E6G mapped within the PB1-binding region; F328L and K340R in the cap-binding region; R663K in the RNA-binding domain. The remaining differences mapped in between those major domains.

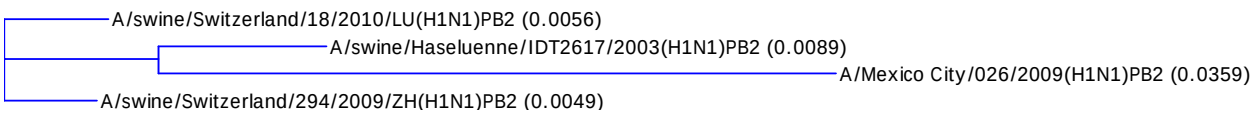

Fig. 24: Guide tree of the alignments of the translated PB2 protein. The predicted PB2 amino acid sequences of 2 Swiss SIV isolates were available for comparison with the pandemic strain Mexico 2009 and the fully sequenced European SIV Haseluenne 2003. Each virus strain is indicated by its full name and the short name for the protein. The calculated distance values are indicated in parenthesis.

Discussion

The present work set out to characterize influenza A viruses (SSIVs) circulating among the Swiss pigs throughout recent years. For this purpose, a collection of one laboratory strain (PR8) and 49 field samples was used, which comprised individual, pre-confirmed SSIV isolates from all over Switzerland, and which had been collected between 2004 and 2010. After optimizing the recently published multiple-segment RT-PCR (MSRT-PCR; 2) for our purpose, the nucleic acids extracted from those 50 samples were subjected to MSRT-PCR and emerging bands were cloned by the TOPO-TA technique prior to sequencing. 44 of the isolates provided at least 1 to maximum 8 bands upon gel electrophoresis of the PCR products, with band sizes consistent with amplification of various influenza virus segments. In contrast, no bands could be achieved from 5 isolates. Cloned bands were subjected to sequencing and, thus, a total of 55 sequences were obtained. Not all viral genome segments were equally represented among the sequences but a full set of genomic sequence was obtained from one isolate (SSIV 18/10) and, accordingly, one major purpose of the present work had been achieved. However, it has to be noted that MSRT-PCR did not amplify all segments equally well. In general, smaller bands were more easily amplified than larger bands. Indeed, it was necessary to make use of a more specific RT-PCR to successfully amplify the three largest genomic segments 1 through 3 of the SSIVs (40). Each cloned PCR product was sequenced in both orientations. Wherever possible, the resulting sequences were aligned to each other, thus, helping to determine the true sequence of each viral isolate. Two types of difficulty were met through this analysis: (1) the very far 3'-ends of some sequences were sometimes difficult to read; (2) in some instances, the consensus sequence would have caused an early translation termination in an essential gene product, a fact that would not be compatible with a viable virus. In such cases, we manually corrected the sequence according to the sequence of its closest neighbor, making use of either other SSIVs or the Haseluenne strain.

Using this approach, 22 sequences were obtained from 9 strains that had been propagated in cell culture, whereas 33 sequences were derived from 6 nasal swabs containing uncultured virus. Thus, our sequencing results partially covered a total of 15 different isolates.

In a second step, these sequences were analyzed by the BLASTn method, which provided a rough estimate of the relation of the novel sequences with those deposited in GeneBank. Obviously, a close relationship with other European SIV isolates was noted but only few of the emerging isolates had been fully sequenced. The one strain that emerged always as very closely related to our SSIVs and one of which a full genomic sequence was available was Influenza A/sw/Haseluenne/2003/H1N1 (48). Therefore, this strain was generally used for comparison with our sequences. Since the origin of the H1N1 pandemic in 2009 and 2010 had been blamed on pigs, it was also of interest to compare our SSIV-derived sequences to the novel pandemic virus (49).

Accordingly, all of our sequences were segment-wise aligned to each other as well as to H1N1 Haseluenne 2003 and the pandemic H1N1 2009. Moreover, the predicted aa sequences of the translated open reading frames encoded in the individual segments were also aligned. It was noted that in all instances the SSIVs were more closely related among each other (90 to >97% identity on the nt level) than to the European SIV Haseluenne 2003 (85 to 95% identity) and all of them were again clearly distinct from the pandemic H1N1 virus of 2009 (<85% identity). The most highly conserved segment was number 7 (88% when Haseluenne and the pandemic H1N1 were included), whereas the highest variability was found in segment 4 (<65% identity). On the amino acid level, the most conserved protein was M1 (>97% identity) and the least conserved was HA with more than 119 aa substitutions between SSIVs and the pandemic H1N1 as compared to <18 substitutions when only the SSIVs were compared against each other. Thus, the Swiss SIVs seemed to constitute their own cluster and are only very distantly related to the pandemic H1N1 virus of 2009. There was absolutely no evidence that a virus similar to the pandemic one did circulate among Swiss pigs, at least not prior to 2011. Thus, a second major aim of our study could be completed. Interestingly, several months after completion of this work, the pandemic H1N1 virus was indeed detected among Swiss pigs with influenza symptoms. Indeed, until October of 2011 the number of confirmed cases of pandemic H1N1 in Swiss pigs increased to 5 (Engels, Heidemeyer, Ackermann, unpublished data). Consequently, one may conclude that the pandemic H1N1 virus has indeed zoonotic potential, although, according to our data, the virus was transmitted first from humans to pigs. It will be interesting to observe, whether or not the new virus will be able to conquer its ecological niche among the Swiss pig population.

The salient features learned from the sequence analysis of the individual segments and their predicted translations can be summarized as follows.

Segment 1. This segment encodes for the viral polymerase subunit PB2 and the Swiss sequences were, as expected, highly conserved with a consensus among the SSIVs of 100%. Four domains have been assigned within the PB2 protein, i.e. the PB1-binding region (aa1 to 35), the cap-binding region (aa318 to 483), the RNA-binding domain (aa 535 to 684) and the nuclear import domain (aa 684 to the C-terminal end) (47). While the nuclear import domain was essentially conserved between the isolates, one amino acid exchange was mapped to within the PB1-binding domain (E6G), and each two to the cap-binding region (F328L and K340R) and the RNA-binding domain (I649V and R663K), respectively.

17 positions have been identified within PB2, which can indicate an av-av or hu-hu transmission (43). Ten of those locations were indicative for av-av transmission in our Swiss isolates as well as Haslunne 2003 and Mexico 2009. However, at position 44, our two SSIVs encoded for Serine, which suggests hu-hu transmission, instead of 44A, which is typical for av-av. Similarly, alanine at position 661 of the two SSIVs changed the sequence to a hu-hu-signature (T in Mexico and Haseluenne, indicative for av-av). D701N, which was detected in both of the SSIVs and in Haseluenne (2003) but not in Mexico (2009) has been reported to be indicative for better replication in human cells and for interaction with importin alpha (50).

Segment 2. This bicistronic segment encodes for the viral polymerase subunit PB1 and for the short proapoptotic peptide PB1-F2 (47). While the entire sequences were very well conserved with 100% consensus on the predicted aa level of the PB1 ORF, a most interesting difference was observed from within the PB1-F2 ORF, which was maintained at a coding capacity of 90 aa in two of the three SSIVs as well as Haseluenne 2003 but early terminated to non-existence in Mexico 2009. The same ORF was truncated to 79 aa

in the third SSIV (247/2009/AG), thus, setting all of the Swiss isolates well apart from the pandemic strain from Mexico. Moreover, all Swiss isolates had most of the predicted key features of PB1-F2 conserved, i.e. W9 and T13 (alpha 1 domain); I16 and K20 (alpha 2 domain); K53, S84, E87 (alpha 3 domain) (46). Only the reported V50 was replaced by D50 in two of the SSIVs (294/2009/ZH and 18/2010/LU) and by G50 in Haseluenne (2003) and SSIV 247/2009/AG.

Segment 3. This segment harbors one single ORF, which translates to PA, one of the three viral polymerase subunits, and encodes for the cap-snatching endonuclease function of the influenzavirus. (51). Both the nucleotide and the predicted aa sequences of the SSIVs were well conserved and clearly distinct from those of Mexico 2009. Similar to Haseluenne (2003), the predicted PA molecule of the SSIVs exceeded the size of the same molecule in Mexico (2009) by four amino acids at the C-terminal end. Although some differences were noticed in the amino terminal sequence 1 to 209, where the endonuclease activity of the molecule has been mapped, the five key active sites were entirely conserved (H41 in alpha helix 3; E80 in beta sheet 1; D108 in beta 2; E119 in beta 3; K134 in alpha 5). Two interesting sites of discrepancy were, however noticed: A20, residing in alpha helix 2, was replaced by T20 in two SSIVs and by I20 in one Swiss isolate. Previously, T20 had been detected in Influenza B virus and I20 in influenza C virus (51).

Segment 4. Segment 4 encodes for the influenzavirus hemagglutinin (HA), the most significant target for immunoprotection. Importantly, all of the Swiss isolates belonged clearly to the H1 subgroup and not a single case of H3 could be detected. This was somewhat surprising, since H3 viruses are known to circulate among European pigs (48). An interesting feature was observed at position 190 of HA, where all strains compared in this study encoded for a Valin (V). Aspartic acid (D) at position 190 has been described as marker for the ability to make use of the human receptor, whereas glutamic acid (E) at this position is favored by viruses using the avian receptor (52, 53, 54). Thus, according to this marker the SSIVs seemed not to be strongly committed to either human-human or avian-avian transmission.

Segment 5. Segment 5 encodes for the multifunctional influenzavirus nucleoprotein (NP), whose primary purpose is to encapsidate the viral genome (46). Interestingly, the Swiss isolates grouped into two distinct branches, one representing strains from Zurich and Lucerne, the other representing isolates from Aargau and Solothurn. Haseluenne and Mexico were positioned on an early subbranch originating from the branch with the Zurich and Lucerne isolates. However, Mexico 2009 was clearly more distinct from the SSIV (222 differences against the consensus) than the Haseluenne 2003 isolate (30 differences). Most of those differences were distributed randomly along the sequence. However, eight loci were detected, which were important for the grouping of the individual isolates on the guide tree. At each one of those positions, one group of the SSIVs shared identity with either Mexico 2009 or Haseluenne 2003, whereas the other group shared with the alternative isolate. However, none of the SSIVs did share constantly with always the same foreign isolate, which suggests that neither of those strains had a decisive role in the parenting of the SSIVs. On the predicted aa level, all compared isolates were highly conserved in their NLS-1 and NLS-2 domains. However, a single aa exchange V329I was observed in the cytoplasmic accumulation signal (CAS) of strain Haseluenne 2003, which was not conserved among the Swiss isolates. Position 366 of NP is known to be important for immune protection against influenza viruses. An aspartic acid (Asp, D) at this position confers a dominant immune protective CD8 epitope (aa 366 to 374), while variations at this position correlate to lower cross

protection and lower CD8 T cell response(64). Interestingly, all of our SSIV strains were predicted to encode for an Alanine at this position.

Segment 6. Segment 6 encodes for the neuraminidase protein (N or NA), which is currently the most important target for antiviral treatment (45). As expected, some variability was noted but the Swiss isolates clustered more closely to each other than to either Haseluenne 2003 or Mexico 2009. Typical domains of the neuraminidase molecule were well conserved, i.e. the short cytoplasmic tail (aa1-6), the transmembrane region (residues 7-34), the asp box motif (aa258-264) and the general boundaries of the sialidase domain (residues 102-448) (55). Similarly, typical posttranslational modification sites were conserved, for example C49 (intermolecular disulfide bridging or palmitoylation site) as well as N146 and N236 (conserved glycosylation sites). The predicted exchange of polar amino acid T20 and S21 by non-polar substitutes A20 and G21, respectively, in the NA aa sequences of some of the SSIVs (18/2010, 206/2009, 270/2009, 294/2009) may be relevant, since they may affect the transmembrane region. Importantly, all of the isolates analyzed in this set of comparison, including Haseluenne and Mexico, featured the R194G mutation, which is considered a prerequisite for the consecutive H274Y mutation (45) Indeed H274Y, which confers Oseltamivir resistance, was also observed in all of our SSIV isolates.

Segment 7. In segment 7, the two expected open reading frames, M1 (252 codons) and M2 (97 codons), which slightly overlap, were well conserved among all the SSIVs as well as Haseluenne 2003 and Mexico 2009. An identity of 98 to 100% was observed on the aa sequence level of M1, which is consistent with the notion that this protein is most highly conserved among influenza A viruses (28). Interestingly, one exception was noted, namely with isolate SIV237/09, which was almost identical to all the other isolates, except for aa 82 to 89 (KCPKRKWG), where it did not match at all to NALNGNGD, which was present in all other predicted sequences. The significance of this difference remains obscure since no other M1 sequences in the databases display similarity to this small stretch in isolate 237, whereas the NALNGNGD sequence is highly conserved. Two Swiss isolates, SIV 237/09 and 270/09 (SG) had exchanged the otherwise conserved Leucine at position 103 for Isoleucine. This mutation may be relevant, since it localizes in the RNA-binding- and nuclear localization domain (56, 57, 58, 59). Another potentially relevant mutation was detected in SIV 226/2009 (TG), where the zinc-binding motif CATCEQIADSQHRSH (aa148 to 162) (56, 60, 61) was affected at position 150 with the replacement of a polar Threonine by a nonpolar Isoleucine (T150I).

Little though decisive variation was detected among the predicted aa sequences of the M2-protein, which is important for viral uncoating and in this context a target for the antiviral compound Amantadine. All of the strains analyzed here coded for an Asparagine (N) at position 31, which confers resistance to Amantadine (62). Interestingly, the Mexico 2009 strain distinguishes itself from the other isolates by coding for a Valine at position 27 of M2, which separately confers resistance against Amantadine (62).

Segment 8. This segment encodes two open reading frames in the negative sense orientation (NS1 and NS2) as well as for an ORF in the opposite sense orientation (NSP). On the nucleotide sequence level, the SSIVs were clearly distinct from Haseluenne 2003 and even more so from Mexico 2009. On the amino acid level, all SSIV sequences showed an identical truncation of NS1 at their C-termini, which disrupted one of two predicted nuclear localization signals from within NS1 (NLS-2)(63). In contrast to the SSIVs, strain Haseluenne featured a normal size NS1 protein with 230 aa (63). The pandemic H1N1 (both Mexico 2009 and Germany 2009) had also a truncated NS1, although with 219 aa. In contrast to NSL-2, the predicted NSL-1 site (aa 30 to 41) was well conserved among

the SSIVs. However, L36 in Haseluenne and the pandemic H1N1 was replaced by isoleucine in all Swiss isolates. Yet, Haseluenne differed at position 39 (E/D) relative to pandemic H1N1 and all SSIVs (63).
The alignment of the predicted NS2 aa sequences revealed only few differences compared to either Haseluenne or pandemic H1N1. The highest variation was seen at positions 22 (G22R) and 23 (S23P).

Conclusions

Overall, our analyses indicate that the SIV strains circulating in Switzerland prior to 2011 were clearly distinct from the pandemic H1N1, which had been observed since 2009 to circulate among the Swiss human population. Only now, this strain is entering the pig population, suggesting that this is actually a zoonotic virus, though, in the case of Switzerland, not originating from pigs and transmitted to humans but rather contrary, originating in humans and transmitted to pigs. It will be of interest to observe, whether or not this new virus will be able to replace the innate virus strains described in the present study. A second interesting feature was that the Swiss SIV isolates seemed to be closely related among each other and distinct from conventional European SIV strains. However, all isolates, of which the neuraminidase aa sequence could be determined, were observed to encode for the H274Y mutation in NA and, thus, were predicted as resistant against Oseltamivir. Moreover, all isolates also featured the R194G mutation in NA, which is considered a prerequisite for the consecutive H274Y mutation (45). Since broad use of Oseltamivir in Swiss pigs has not been reported, the question arises as to how these consecutive mutations have emerged among the SSIVs. One possible answer might be that the corresponding loci are just prone for these mutations and, thus, have acquired the same mutations at the same time as independent virus strains outside of Switzerland. Yet, how likely could that be in reality? A second theory might argue that these viruses represent descendants from Oseltamivir-resistant human viruses that have acquired the same mutations while circulating among humans. However, our sequencing results speak against this latter theory. A third explanation may be the notion that our Swiss pig population is actually not as protected from SIVs circulating in Europe as we like to believe and as it is suggested by the viral sequences that seem to distinguish themselves from other European isolates.

Appendices

Appendix 1a

Segment 8

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SSIV 199/09 NS      ACGCGTGATCAGCAAAAGCAGGG-TGACAAAAACATAATGGATTCAAATA
SSIV 132/09 NS      ACGCGTGATCAGCAAAAGCAGGGGTGACAAAAACATAATGGATTCAAATA
SSIV 247/09 NS      ACGCGTGATCAGCAAAAGCAGGG-TGACAAAAACATAATGGATTCAAATA
SSIV 45/10 NS       ACGCGTGATCAGCAAAAGCAGGGGTGACAAAAACATAATGGATTCAAATA
SSIV 54/10 NS       ACGCGTGATCAGCAAAAGCAGGGGTGACAAAAACATAATGGATTCAAATA
SSIV 88/09 NS       ACGCGTGATCAGCAAAAGCAGGG-TGACAAAAACATAATGGATTCAAATA
SSIV 206/09 NS      ACGCGTGATCAGCAAAAGCAGGG-TGACAAAAACATAATGGATTCAAATA
SSIV 270/09 NS      ACGCGTGATCAGCAAAAGCAGGG-TGACAAAAACATAATGGATTCAAATA
SSIV 18/10 NS       ACGCGTGATCAGCAAAAGCAGGG-TGACAAAAACATAANGGATTCAAATA
SSIV 237/09 NS      ACGCGTGATCAGCAAAAGCAGGG-TGACAAAAACATAATGGATTCAAATA
SSIV 157/09 NS      --GCGTGATCAGCAAAAGCAGGG-TGNCAAAAACATAATGGATTCAAATA
SSIV 294/09 NS      --GCGTGATCAGCAAAGCAGGGG-TGACAAAAACATAATGGATTCAAATA
SSIV 54/04 NS       ACGCGTGATCAGCAAAAGAAGGG-TGACAAAAACATAATGGATTCAAATA
Haseluenne 2003     ----------AGCAAAAGCAGGG-TGACAAAAACATGATGGATTCAAACA
Mexico 2009         -------------------------------AACATAATGGACTCCAACA
                                                   ***** * *** ** ** *

SSIV 199/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTTCTTTGGCATGTCCGCAAACGA
SSIV 132/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTTCTTTGGCATGTCCGCAAACGA
SSIV 247/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTCCTTTGGCATGTCCGCAAACGA
SSIV 45/10 NS       CTGTGTCAAGCTTTCAGGTAGACTGCTTTCTTTGGCATGTCCGCAAACGA
SSIV 54/10 NS       CTGTGTCAAGCTTTCAGGTAGACTGCTTTCTTTGGCATGTCCGCAAACGA
SSIV 88/09 NS       CTGTGTCAAGCTTTCAGGTAGACTGCTTTCTTTGGCATGTCCGCAAACGA
SSIV 206/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTCCTTTGGCATGTCCGCAAACGA
SSIV 270/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTCCTTTGGCATGTCCGCAAACGA
SSIV 18/10 NS       CTGTGTCAAGCTTTCAGGTAGACTGCTTCCTTTGGCATGTCCGCAAACGA
SSIV 237/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTCCTTTGGCATGTCCGCAAACGA
SSIV 157/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTCCTTTGGCATGTCCGCAAACGA
SSIV 294/09 NS      CTGTGTCAAGCTTTCAGGTAGACTGCTTCCTTTGGCATGTCCGCAAACGA
SSIV 54/04 NS       CTGTGTCAAGCTTTCAGGTAGACTGTTTTCTTTGGCATGTCCGCAAACGA
Haseluenne 2003     CTGTGTCAAGCTTTCAGGTAGACTGCTTTCTTTGGCATGTCCGCAAACGG
Mexico 2009         CCATGTCAAGCTTTCAGGTAGACTGTTTCCTTTGGCATATCCGCAAGCGA
                    *  ********************** ** ********* ******* **

SSIV 199/09 NS      TTTGCAGACTGTGGGCTTAGTGATGCACCATTTCTTGATAGGATTCGCCG
SSIV 132/09 NS      TTTGCAGACTGTGGGCTTAGTGATGCACCATTTCTTGATAGGATTCGCCG
SSIV 247/09 NS      TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 45/10 NS       TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 54/10 NS       TTTGCAGACTGTGGGCTTGGTGATGNNC-GTTTCTTGGTAGGATTCGCCG
SSIV 88/09 NS       TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 206/09 NS      TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 270/09 NS      TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 18/10 NS       TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 237/09 NS      TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 157/09 NS      TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 294/09 NS      TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
SSIV 54/04 NS       TTTGCAGACTGTGGGCTTGGTGATGCCCCATTTCTTGATAGGATTCGCCG
Haseluenne 2003     TTTGCAGACTGGGGGCTTGGCGATGCCCCATTTCTCGATAGGCTTCGCCG
Mexico 2009         TTTGCAGACAATGGATTGGGTGATGCCCCATTCCTTGATCGGCTCCGCCG
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SSIV 199/09 NS      AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATCG
SSIV 132/09 NS      AGATCAAAAGTCCCTAAGGGGGAGAGGCAACACTCTTGGTCTGGAAATCG
SSIV 247/09 NS      AGATCAAAAGTCCCTAANNG-AAGAGGCAACACTCTTGGTCTGGAAATCG
SSIV 45/10 NS       AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATCG
SSIV 54/10 NS       AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATCG
SSIV 88/09 NS       AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATCG
SSIV 206/09 NS      AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATTG
SSIV 270/09 NS      AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATTG
SSIV 18/10 NS       AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATTG
SSIV 237/09 NS      AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATTG
SSIV 157/09 NS      AGATCAAAAATCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATTG
SSIV 294/09 NS      AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATTG
SSIV 54/04 NS       AGATCAAAAGTCCCTAAGGGGAAGAGGCAACACTCTTGGTCTGGAAATCG
Haseluenne 2003     AGACCAGAAGTCCCTAAGAGGGAGAGGCAGCACTCTTGGTCTGGAAATCG
Mexico 2009         AGATCAAAAGTCCTTAAAAGGAAGAGGCAACACCCTTGGCCTCGATATCG
                    *** ** ** *** ***  *  ******* *** ***** ** ** ** *

SSIV 199/09 NS      AACCAGCCACTCGTGCTGGGAAACAAATAGTGAAGCGTATCCTGGGAGAA
SSIV 132/09 NS      AACCAGCCACTCGTGCTGGGAAACAAATAGTGAAGCGTATCCTGGGAGAA
SSIV 247/09 NS      AACCAGCCACTCGTGCTGGGAAACAAATAGTGAAGCGTATCCTGGGAGAA
SSIV 45/10 NS       AACCAGCCACTCGTGCTGGGGAACAAATAGTGAAGCGTATCCTAGGAGAA
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SSIV 54/10 NS       AACCAGCCACTCGTGCTGGGAAACAAATAGTGAAGCGCATCCTAGGAGAA
SSIV 88/09 NS       AACCAGCCACTCGTGCTGGGAAACAAATAGTGAAGCGTATCCTAGGAGAA
SSIV 206/09 NS      AACCAGCCACTCATGCTGGGAAACAAATAGTGAAGCGTATCCTAGAAGAA
SSIV 270/09 NS      AACCAGCCACTCATGCTGGGAAACAAATAGTGAAGCGTATCCTAGAAGAA
SSIV 18/10 NS       AATCAGCCACTCATGCTGGGAAACAAATAGTGAAGCGTATCCTAGAAGAA
SSIV 237/09 NS      AACCAGCCACTCATGCTGGGAAACAAATAGTGAAGCGTATCCTAGAAGAA
SSIV 157/09 NS      AACCAGCCACTCATGCTGGGAAACAAATAGTGAAGCGTATCCTAGAAGAA
SSIV 294/09 NS      AACCAGCCACTCATGCTGGGAAACAAATAGTGAAGCGTATCCTAGAAGAA
SSIV 54/04 NS       AACCAGCCACTCGTGCTGGGAAACAAATAGTGAAGCGTATCCTAGAAGAA
Haseluenne 2003     AACCAGCTACTCGTGCGGGGAAACAAATAGTGAAGCGTATTCTGGAAGAA
Mexico 2009         AAACAGCCACTCTTGTTGGGAAACAAATCGTGGAATGGATCTTGAAAGAG
                    ** **** **** **  *** ******* *** *  * **  *   ***

SSIV 199/09 NS      GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCCACTTCACG
SSIV 132/09 NS      GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCCACTTCACG
SSIV 247/09 NS      GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCTACTTCACG
SSIV 45/10 NS       GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCTACTTCACG
SSIV 54/10 NS       GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCTACTTCACG
SSIV 88/09 NS       GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCTACTTCACG
SSIV 206/09 NS      GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCTACTTCACG
SSIV 270/09 NS      GAATACGATGAGGCATTTAAACTAACGATAGCTTCCGTGCCTACTTCACG
SSIV 18/10 NS       GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCTACTTCACG
SSIV 237/09 NS      GAATACGATGAGGCATTTAAACTAACGATAGCTTCCGTGCCTACTTCACG
SSIV 157/09 NS      GAATACGATGAGGCATTTAAACTAATGATAGCTTCTGTGCCTACTTCACG
SSIV 294/09 NS      GAATACGATAAGGCATTTCAACTAACGATAGCTTCTGTGCCTACTTCACG
SSIV 54/04 NS       GAATACGATGAGGCATTTAAACTAACGATAGCTTCTGTGCCTACTTCACG
Haseluenne 2003     GAATACGAGGAGGCACTTAAAATAACTATCGCTTCTGTGCCTGCTTCACG
Mexico 2009         GAATCCAGCGAGACACTTAGAATGACAATTGCATCTGTACCTACTTCGCG
                    **** *    ** ** **  * * *  ** ** ** ** **  **** **

SSIV 199/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGC
SSIV 132/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGC
SSIV 247/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGC
SSIV 45/10 NS       CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGATACTGGTTCATGC
SSIV 54/10 NS       CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGC
SSIV 88/09 NS       CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGC
SSIV 206/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGA
SSIV 270/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGA
SSIV 18/10 NS       CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGA
SSIV 237/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGA
SSIV 157/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGA
SSIV 294/09 NS      CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGA
SSIV 54/04 NS       CTATCTAACTGACATGACTCTTGAAGAAATGTCAAGAGACTGGTTCATGC
Haseluenne 2003     CTATCTAACTGACATGACTCTTGAAGAGATGTCAAGGGACTGGTTCATGC
Mexico 2009         CTACCTTTCTGACATGACCCTCGAGGAAATGTCACGAGACTGGTTCATGC
                    *** **  ********** ** ** ** ****** *  ***********

SSIV 199/09 NS      TCATGCCCAAGCAAAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 132/09 NS      TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 247/09 NS      TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAAAATGGACCAA
SSIV 45/10 NS       TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 54/10 NS       TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 88/09 NS       TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 206/09 NS      TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 270/09 NS      TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 18/10 NS       TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 237/09 NS      TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 157/09 NS      TCATGCCCAAGAAAAAAGTGGCAGGTTCCCTCTGCATCAGAATGGACCAA
SSIV 294/09 NS      TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
SSIV 54/04 NS       TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAGAATGGACCAA
Haseluenne 2003     TCATGCCCAAGCAGAAAGTGGCAGGTTCCCTTTGCATCAAAATGGACCAG
Mexico 2009         TCATGCCTAGGCAAAAGATAATAGGCCCTCTTTGCGTGCGATTGGACCAG
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SSIV 199/09 NS      GCAATAATGAATAAAAAAATCACATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 132/09 NS      GCAATAATGAATAAAAAAATCACATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 247/09 NS      GCAATAATGAATAAAAAAATCACATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 45/10 NS       GCAATAATGAATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 54/10 NS       GCAATAATGAATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 88/09 NS       GCAATAATGAATAAAAAAATCATATTGAAAGCAGACTTCAGTGTGATTTT
SSIV 206/09 NS      GCAATAATGGATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 270/09 NS      GCAATAATGGATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 18/10 NS       GCAATAATGGATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 237/09 NS      GCAATAATGGATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 157/09 NS      GCAATAATGGATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 294/09 NS      GCAATAATGGATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
SSIV 54/04 NS       GCAATAATGGATAAAAAAATCATATTGAAAGCAAACTTCAGTGTGATTTT
Haseluenne 2003     GCAATAATGGATAAATACATCACATTGAAAGCAAACTTCAGTGTGATTTT
Mexico 2009         GCGATCATGGAAAAGAACATAGTACTGAAAGCGAACTTCAGTGTAATCTT
                    ** ** *** * **  * **   * *******  ********** ** **

SSIV 199/09 NS      TGGTCGACTGGAAACCCTGATACTCCTTAGAGCTTTCACGGAAGAAGGGG
SSIV 132/09 NS      TGGTCGACTGGAAACCCTGATACTCCTTAGAGCTTTCACGGAAGAAGGGG
SSIV 247/09 NS      TGGTCGACTGGAAACCCTGATACTCCTTAGAGCTTTCACGGAAGAAGGGG
SSIV 45/10 NS       TGGTCGACTGGAAACCCTAATACTCCTTAGAGCTTTCACGGAAGAAGGGG
SSIV 54/10 NS       TGGTCGACTTGAAACCCTGATACTCCTTAGAGCTTTCACGGAAGAAGGGG
SSIV 88/09 NS       TGATCGACTGGAAACCCTGATACTCCTTAGAGCTTTCACGGAAGAAGGGG
SSIV 206/09 NS      TGATCGACTGGAAACCCTTATACTCCTTAGAGCTTTCACAGAAGAAGGGG
SSIV 270/09 NS      TGATCGACTGGAAACCCTTATACTCCTTAGAGCTTTCACAGAAGAAGGGG
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SSIV 18/10 NS        TGATCGACTGGAAACCCTTATACTCCTTAGAGCTTTCACAGAAGAAGGGG
SSIV 237/09 NS       TGATCGACTGGAAACCCTTATACTCCTTAGAGCTTTCACAGAAGAAGGGG
SSIV 157/09 NS       TGATCGACTGGAAACCCTCATACTCCTTAGAGCTTTCACAGAAGAAGGGG
SSIV 294/09 NS       TGATCGACTGGAAACCCTTATACTCCTTAGAGCTTTCACAGAAGAAGGGG
SSIV 54/04 NS        TGGTCGACTGGAAACCCTGATACTCCTTAGAGCTTTCACGGAAGAAGGGG
Haseluenne 2003      TGATCGACTGGAAACTCTGATACTCCTTAGAGCTTTCACGGAAGAAGGGG
Mexico 2009          TAACCGATTAGAGACCTTGATACTACTAAGGGCTTTCACTGAGGAGGGAG
                     *   *** * ** **  * ***** ** ** ******** ** ** ** *

SSIV 199/09 NS       CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 132/09 NS       CAATTGTAGGTGAAGTCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 247/09 NS       CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 45/10 NS        CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 54/10 NS        CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 88/09 NS        CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 206/09 NS       CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 270/09 NS       CAATTGTAGGTGAAATCCCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 18/10 NS        CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 237/09 NS       CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 157/09 NS       CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 294/09 NS       CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
SSIV 54/04 NS        CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
Haseluenne 2003      CAATTGTAGGTGAAATCTCACCGTTACCTTCTCTTCCAGGACATACTGAT
Mexico 2009          CAATAGTTGGAGAAATTTCACCATTACCTTCTCTTCCAGGACATACTTAT
                     **** ** ** *** *  **** ************************ **

SSIV 199/09 NS       GAGGATGTCAAAGATGCAATTGAGATCCTCATCAAAGGACTTGAATGGAA
SSIV 132/09 NS       GAGGATGTCAAAGATGCAATTGAGATCCTCATCAAAGGACTTGAATGGAA
SSIV 247/09 NS       GAGGATGTCAAAGATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 45/10 NS        GAGGATGTCAAAGATGCAATTGAGACCCTCATCAAAGGACTTGAATGGAA
SSIV 54/10 NS        GAGGATGTCAAAGATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 88/09 NS        GAGGATGTCAAAGATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 206/09 NS       GAGGATGTCAAAAATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 270/09 NS       GAGGATGTCAAAAATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 18/10 NS        GAGGATGTCAAAAATGCAATTGGGATCCTCATCAAAGGACTTAAATGGAA
SSIV 237/09 NS       GAGGATGTCAAAAATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 157/09 NS       GAGGATGTCAAAAATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 294/09 NS       GAGGATGTCAAAAATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
SSIV 54/04 NS        GAGGATGTCAAAAATGCAATTGGGATCCTCATCAAAGGACTTGAATGGAA
Haseluenne 2003      GAGGATGTCAAAAATGCAATTGGGATCCTCATCAGAGGACTTGAATGGAA
Mexico 2009          GAGGATGTCAAAAATGCAGTTGGGGTCCTCATCGGAGGACTTGAATGGAA
                     ************ ***** *** *  *******  ******* *******

SSIV 199/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 132/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 247/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 45/10 NS        TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 54/10 NS        TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 88/09 NS        TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 206/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 270/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 18/10 NS        TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 237/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 157/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 294/09 NS       TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
SSIV 54/04 NS        TGATAACACAGTTCGAGTCTCTGAAGCTCTACAGAGATTCACTTGGAGAA
Haseluenne 2003      TGATAACACAATTCGAGTCTCTGAGGCTCTACAGAGATTCGCTTGGAGAA
Mexico 2009          TGGTAACACGGTTCGAGTCTCTGAAAATATACAGAGATTCGCTTGGAGAA
                     ** ******  *************   * ************ *********

SSIV 199/09 NS       GCACTAATGAGAACGGGGGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 132/09 NS       GCACTAATGAGAACGGGGGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 247/09 NS       GCACTAATGAGAACGGGGGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 45/10 NS        GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 54/10 NS        GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 88/09 NS        GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 206/09 NS       GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 270/09 NS       GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 18/10 NS        GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 237/09 NS       GCACTAATGAGAATGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 157/09 NS       GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 294/09 NS       GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
SSIV 54/04 NS        GCACTAATGAGAACGGGAGACCTCCATTATCTCCAAGATAGAAACGGAAA
Haseluenne 2003      GCACTAATGAGAACGGGAGACCTCCATTTCCTCCAAAACAGAAACGGAAA
Mexico 2009          ACTGTGATGAGAATGGGAGACCTTCACTACCTCCAGAGCAGAAATGAAAA
                      *  * ******* *** ***** ** *  *****    ***** * ***

SSIV 199/09 NS       ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTAATTG
SSIV 132/09 NS       ATGGCGAGAACAATTGGGTCAGAAGTTTGAAGAGATAAGATGGCTAATTG
SSIV 247/09 NS       ATGGCGAGAAAAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTAATTG
SSIV 45/10 NS        ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTAATTG
SSIV 54/10 NS        ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTAATTG
SSIV 88/09 NS        ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAAATAAGATGGCTAATTG
SSIV 206/09 NS       ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTGATTG
SSIV 270/09 NS       ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTGATTG
SSIV 18/10 NS        ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTGATTG
SSIV 237/09 NS       ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTGATTG
SSIV 157/09 NS       ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTGATTG
SSIV 294/09 NS       ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTGATTG
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SSIV 54/04 NS       ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAGATAAGATGGCTGATTG
Haseluenne 2003     ATGGCGAGAACAATTGGGCCAGAAGTTTGAAGAAATAAGATGGCTAATTG
Mexico 2009         GTGGCGAGAGCAATTGGGACAGAAATTTGAGGAAATAAGGTGGTTAATTG
                     ********  ******* ***** ***** ** ***** *** * ****

SSIV 199/09 NS      AAGAAGTGCGACACAGATTAAAATTAACCGAGAATAGCTTTGAGCAGATA
SSIV 132/09 NS      AAGAAGTGCGACACAGATTAAAATTAACCGAGAATAGCTTTGAGCAGATA
SSIV 247/09 NS      AAGAAGTGCGACACAGATTAAAATTAACCGAGAATAGCTTTGAGCAGATA
SSIV 45/10 NS       AAGAAGTGCGACACAGATTAAAATTGACCGAGAATAGCTTTGAGCAGATA
SSIV 54/10 NS       AGGAAGTGCGACACAGATTAAAATTAACCGAGAATAGCTTTGAGCAGATA
SSIV 88/09 NS       AAGAAGTGCGACACAGATTAAAATTAACCGAGAATAGCTTTGAGCAGATA
SSIV 206/09 NS      AAGAAGTGCGACACAGATTAAAATTAACAGAGAATAGCTTTGAGCAGATA
SSIV 270/09 NS      AAGAAGTGCGACACAGATTAAAATTAACAGAGAATAGCTTTGAGCAGATA
SSIV 18/10 NS       AAGAAGTGCGACACAGATTAAAATTAACAGAGAATAGCTTTGAGCAGATA
SSIV 237/09 NS      AAGAAGTGCGACACAGACTAAAATTAACAGAGAATAGCTTTGAGCAGATA
SSIV 157/09 NS      AAGAAGTGCGACACAGATTAAAATTGACAGAAAATAGCTTTGAGCAGATA
SSIV 294/09 NS      AAGAAGTGCGACACAGATTAAAATTAACAGAGAATAGCTTTGAGCAGATA
SSIV 54/04 NS       AAGAAGTGCGACACAGATTAAAATTAACAGAGAATAGCTTTGAGCAGATA
Haseluenne 2003     AAGAAGTGCGGCACAGATTAAAAATAACAGAGAATAGCTTTGAACAGATA
Mexico 2009         AAGAAATGCGGCACAGATTGAAAGCGACAGAGAATAGTTTCGAACAAATA
                    * *** **** ****** * ***   ** ** ***** ** ** ** ***

SSIV 199/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 132/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 247/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 45/10 NS       ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 54/10 NS       ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 88/09 NS       ACTTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 206/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 270/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 18/10 NS       ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 237/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 157/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 294/09 NS      ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
SSIV 54/04 NS       ACGTTCATGCAAGCATTACAACTACTGCTTGAAGTGGAGCAAGAGATAAG
Haseluenne 2003     ACGTTTATGCAAGCATTACAACTACTGCTTGAGGTGGAGCAAGAGATAAG
Mexico 2009         ACATTTATGCAAGCCTTACAACTACTGCTTGAAGTAGAACAAGAGATAAG
                    ** ** ******** ****************** ** ** ***********

SSIV 199/09 NS      GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 132/09 NS      GACATTCTCGTTTCAGTTTATTTAATGATAAAAAAACACCCTTGTTTCTA
SSIV 247/09 NS      GACATTCTCGTTTCAGTTTATTTAATNATAAAAAA-CACCCTTGTTTCTA
SSIV 45/10 NS       GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 54/10 NS       GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 88/09 NS       GACATTTTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 206/09 NS      GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 270/09 NS      GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 18/10 NS       GACATTCTCGTTTCAGTTTATTTAATNATAAAAAA-CACCCTTGTTTCTA
SSIV 237/09 NS      GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 157/09 NS      GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
SSIV 294/09 NS      GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCNA
SSIV 54/04 NS       GACATTCTCGTTTCAGTTTATTTAATGATAAAAAA-CACCCTTGTTTCTA
Haseluenne 2003     GACTTTCTCGTTTCAGTTTATTTAATAATAAAAAA-CACCC---------
Mexico 2009         AGCTTTCTCGTTTCAGCTTATTTAATGATAA-------------------
                    * ** ********* ********* ****

SSIV 199/09 NS      CTGATCACGCGT
SSIV 132/09 NS      CTGATCACGCGT
SSIV 247/09 NS      CTGATCACGCGA
SSIV 45/10 NS       CTGATCACGCGT
SSIV 54/10 NS       CTGATCACGCGT
SSIV 88/09 NS       CTGATCACGCGT
SSIV 206/09 NS      CK-ATCACGCGT
SSIV 270/09 NS      CTKATCACGCGT
SSIV 18/10 NS       CN-ATCACGCGT
SSIV 237/09 NS      CNGATCACGCGT
SSIV 157/09 NS      CNGATCACGCGT
SSIV 294/09 NS      CTGATCACGCGT
SSIV 54/04 NS       CTGATCACGCGT
Haseluenne 2003     ------------
Mexico 2009         ------------
```

Legend:
Complete nucleotide sequences of segment 8. Highlighted in blue: mutation for L36I in the translated amino acid sequence. Highlighted in purple: position 70G in in the translated amino acid sequence of NS2 protein; indicator for hu-hu transmission. The STOP-codon, which leads to a shortened NS1 protein, highlighted in red.

Appendix 1b

NS1 protein

```
SSIV 132/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLSDAPFLDRIRRDQKSLRGRGNTL
SSIV 199/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLSDAPFLDRIRRDQKSLRGRGNTL
SSIV 247/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 45/10 NS1    MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 54/10 NS1    MDSNTVSSFQVDCFLWHVRKRFADCGLGDGAFLGRIRRDQKSLRGRGNTL
SSIV 88/09 NS1    MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 54/04 NS1    MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 206/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 237/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 270/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 157/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 294/09 NS1   MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
SSIV 18/10 NS1    MDSNTVSSFQVDCFLWHVRKRFADCGLGDAPFLDRIRRDQKSLRGRGNTL
Haseluenne 2003   MDSNTVSSFQVDCFLWHVRKRFADWGLGDAPFLDRLRRDQKSLRGRGSTL
Mexico 2009       MDSNTMSSFQVDCFLWHIRKRFADNGLGDAPFLDRLRRDQKSLKGRGNTL
                  *****:***********:****** **.*..**.*:*******:***.**

SSIV 132/09 NS1   GLEIEPATRAGKQIVKRILGEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 199/09 NS1   GLEIEPATRAGKQIVKRILGEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 247/09 NS1   GLEIEPATRAGKQIVKRILGEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 45/10 NS1    GLEIEPATRAGEQIVKRILGEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 54/10 NS1    GLEIEPATRAGKQIVKRILGEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 88/09 NS1    GLEIEPATRAGKQIVKRILGEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 54/04 NS1    GLEIEPATRAGKQIVKRILEEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 206/09 NS1   GLEIEPATHAGKQIVKRILEEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 237/09 NS1   GLEIEPATHAGKQIVKRILEEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 270/09 NS1   GLEIEPATHAGKQIVKRILEEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
SSIV 157/09 NS1   GLEIEPATHAGKQIVKRILEEEYDEAFKLMIASVPTSRYLTDMTLEEMSR
SSIV 294/09 NS1   GLEIEPATHAGKQIVKRILEEEYDKAFQLTIASVPTSRYLTDMTLEEMSR
SSIV 18/10 NS1    GLEIESATHAGKQIVKRILEEEYDEAFKLTIASVPTSRYLTDMTLEEMSR
Haseluenne 2003   GLEIEPATRAGKQIVKRILEEEYEEALKITIASVPASRYLTDMTLEEMSR
Mexico 2009       GLDIETATLVGKQIVEWILKEESSETLRMTIASVPTSRYLSDMTLEEMSR
                  **:**.** .*:***: ** ** .:::::: *****:****:*********

SSIV 132/09 NS1   DWFMLMPKQKVAGSLCIRMDQAIMNKKITLKANFSVIFGRLETLILLRAF
SSIV 199/09 NS1   DWFMLMPKQKVAGSLCIRMDQAIMNKKITLKANFSVIFGRLETLILLRAF
SSIV 247/09 NS1   DWFMLMPKQKVAGSLCIKMDQAIMNKKITLKANFSVIFGRLETLILLRAF
SSIV 45/10 NS1    YWFMLMPKQKVAGSLCIRMDQAIMNKKIILKANFSVIFGRLETLILLRAF
SSIV 54/10 NS1    DWFMLMPKQKVAGSLCIRMDQAIMNKKIILKANFSVIFGRLETLILLRAF
SSIV 88/09 NS1    DWFMLMPKQKVAGSLCIRMDQAIMNKKIILKADFSVIFDRLETLILLRAF
SSIV 54/04 NS1    DWFMLMPKQKVAGSLCIRMDQAIMDKKIILKANFSVIFGRLETLILLRAF
SSIV 206/09 NS1   DWFMIMPKQKVAGSLCIRMDQAIMDKKIILKANFSVIFDRLETLILLRAF
SSIV 237/09 NS1   DWFMIMPKQKVAGSLCIRMDQAIMDKKIILKANFSVIFDRLETLILLRAF
SSIV 270/09 NS1   DWFMIMPKQKVAGSLCIRMDQAIMDKKIILKANFSVIFDRLETLILLRAF
SSIV 157/09 NS1   DWFMIMPKKKVAGSLCIRMDQAIMDKKIILKANFSVIFDRLETLILLRAF
SSIV 294/09 NS1   DWFMIMPKQKVAGSLCIRMDQAIMDKKIILKANFSVIFDRLETLILLRAF
SSIV 18/10 NS1    DWFMIMPKQKVAGSLCIRMDQAIMDKKIILKANFSVIFDRLETLILLRAF
Haseluenne 2003   DWFMLMPKQKVAGSLCIKMDQAIMDKYITLKANFSVIFDRLETLILLRAF
Mexico 2009       DWFMLMPRQKIIGPLCVRLDQAIMEKNIVLKANFSVIFNRLETLILLRAF
                  ***:**::*: *.**:::*****:* * ***:*****.***********

SSIV 132/09 NS1   TEEGAIVGEVSPLPSLPGHTDEDVKDAIEILIKGLEWNDNTVRVSEALQR
SSIV 199/09 NS1   TEEGAIVGEISPLPSLPGHTDEDVKDAIEILIKGLEWNDNTVRVSEALQR
SSIV 247/09 NS1   TEEGAIVGEISPLPSLPGHTDEDVKDAIGILIKGLEWNDNTVRVSEALQR
SSIV 45/10 NS1    TEEGAIVGEISPLPSLPGHTDEDVKDAIETLIKGLEWNDNTVRVSEALQR
SSIV 54/10 NS1    TEEGAIVGEISPLPSLPGHTDEDVKDAIGILIKGLEWNDNTVRVSEALQR
SSIV 88/09 NS1    TEEGAIVGEISPLPSLPGHTDEDVKDAIGILIKGLEWNDNTVRVSEALQR
SSIV 54/04 NS1    TEEGAIVGEISPLPSLPGHTDEDVKNAIGILIKGLEWNDNTVRVSEALQR
SSIV 206/09 NS1   TEEGAIVGEISPLPSLPGHTDEDVKNAIGILIKGLEWNDNTVRVSEALQR
SSIV 237/09 NS1   TEEGAIVGEISPLPSLPGHTDEDVKNAIGILIKGLEWNDNTVRVSEALQR
SSIV 270/09 NS1   TEEGAIVGEIPPLPSLPGHTDEDVKNAIGILIKGLEWNDNTVRVSEALQR
SSIV 157/09 NS1   TEEGAIVGEISPLPSLPGHTDEDVKNAIGILIKGLEWNDNTVRVSEALQR
SSIV 294/09 NS1   TEEGAIVGEISPLPSLPGHTDEDVKNAIGILIKGLEWNDNTVRVSEALQR
SSIV 18/10 NS1    TEEGAIVGEISPLPSLPGHTDEDVKNAIGILIKGLKWNDNTVRVSEALQR
Haseluenne 2003   TEEGAIVGEISPLPSLPGHTDEDVKNAIGILIRGLEWNDNTIRVSEALQR
Mexico 2009       TEEGAIVGEISPLPSLPGHTYEDVKNAVGVLIGGLEWNGNTVRVSENIQR
                  *********:.********* ****:*:  ** **:**.**:**** :**

SSIV 132/09 NS1   FTWRSTNENGGPPLSPR-------------
SSIV 199/09 NS1   FTWRSTNENGGPPLSPR-------------
SSIV 247/09 NS1   FTWRSTNENGGPPLSPR-------------
SSIV 45/10 NS1    FTWRSTNENGRPPLSPR-------------
SSIV 54/10 NS1    FTWRSTNENGRPPLSPR-------------
SSIV 88/09 NS1    FTWRSTNENGRPPLSPR-------------
SSIV 54/04 NS1    FTWRSTNENGRPPLSPR-------------
SSIV 206/09 NS1   FTWRSTNENGRPPLSPR-------------
SSIV 237/09 NS1   FTWRSTNENGRPPLSPR-------------
SSIV 270/09 NS1   FTWRSTNENGRPPLSPR-------------
SSIV 157/09 NS1   FTWRSTNENGRPPLSPR-------------
SSIV 294/09 NS1   FTWRSTNENGRPPLSPR-------------
SSIV 18/10 NS1    FTWRSTNENGRPPLSPR-------------
Haseluenne 2003   FAWRSTNENGRPPFPPKQKRKMARTIGPEV
Mexico 2009       FAWRNCDENGRPSLPPEQK-----------
                    *:**. :*** *.:.*.
```

Legend:
Amino acid sequences of NS1 protein. Highlighted in blue: Mutation L36I, which is typical for Swiss Swine influenza isolates. Highlighted in red:

missing amino acids in the Swiss strains, which deletes part of NLS II (218-230).

Appendix 1c

NS2 protein

```
SSIV 132/09 NS2   MDSNTVSSFQDILMRMSKMQLRSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 199/09 NS2   MDSNTVSSFQDILMRMSKMQLRSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 45/10 NS2    MDSNTVSSFQDILMRMSKMQLRPSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 54/10 NS2    MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 88/09 NS2    MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 54/09 NS2    MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 294/09 NS2   MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 270/09 NS2   MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 206/09 NS2   MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 18/10 NS2    MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 157/09 NS2   MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 247/09 NS2   MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
SSIV 237/09 NS2   MDSNTVSSFQDILMRMSKMQLGSSSKDLNGMITQFESLKLYRDSLGEALM
Haseluenne 2003   MDSNTVSSFQDILMRMSKMQLGSSSEDLNGMITQFESLRLYRDSLGEALM
Mexico 2009       MDSNTMSSFQDILMRMSKMQLGSSSEDLNGMVTRFESLKIYRDSLGETVM
                  *****:*************** .**:*****:*:****::*******::*

SSIV 132/09 NS2   RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 199/09 NS2   RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 45/10 NS2    RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 54/10 NS2    RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 88/09 NS2    RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 54/09 NS2    RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 294/09 NS2   RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 270/09 NS2   RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 206/09 NS2   RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 18/10 NS2    RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 157/09 NS2   RTGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 247/09 NS2   RTGDLHYLQDRNGKWREKLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
SSIV 237/09 NS2   RMGDLHYLQDRNGKWREQLGQKFEEIRWLIEEVRHRLKLTENSFEQITFM
Haseluenne 2003   RTGDLHFLQNRNGKWREQLGQKFEEIRWLIEEVRHRLKITENSFEQITFM
Mexico 2009       RMGDLHYLQSRNEKWREQLGQKFEEIRWLIEEMRHRLKATENSFEQITFM
                  ****:**.** ****:**************:***** ***********

SSIV 132/09 NS2   QALQLLLEVEQEIRTFSFQFI
SSIV 199/09 NS2   QALQLLLEVEQEIRTFSFQFI
SSIV 45/10 NS2    QALQLLLEVEQEIRTFSFQFI
SSIV 54/10 NS2    QALQLLLEVEQEIRTFSFQFI
SSIV 88/09 NS2    QALQLLLEVEQEIRTFSFQFI
SSIV 54/09 NS2    QALQLLLEVEQEIRTFSFQFI
SSIV 294/09 NS2   QALQLLLEVEQEIRTFSFQFI
SSIV 270/09 NS2   QALQLLLEVEQEIRTFSFQFI
SSIV 206/09 NS2   QALQLLLEVEQEIRTFSFQFI
SSIV 18/10 NS2    QALQLLLEVEQEIRTFSFQFI
SSIV 157/09 NS2   QALQLLLEVEQEIRTFSFQFI
SSIV 247/09 NS2   QALQLLLEVEQEIRTFSFQFI
SSIV 237/09 NS2   QALQLLLEVEQEIRTFSFQFI
Haseluenne 2003   QALQLLLEVEQEIRTFSFQFI
Mexico 2009       QALQLLLEVEQEIRAFSFQLI
                  **************:****:*
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Legend:
Amino acid sequences of NS2 protein. Purple: 70G, indicator for hu-hu transmission.

Appendix 2a

Segment 7

```
SSIV 54/10 M       ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 132/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 45/10 M       ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 270/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 237/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 294/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 247/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 18/10 M       ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 206/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 226/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 25/06 M       ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 54/04 M       ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
SSIV 157/09 M      ACGCGTGATCAGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
Haseluenne 2003    ----------AGCAAAAGCAGGTAGATATTTAAAGATGAGTCTTCTAACC
AMexico 2009       ------------------------------TAAAGATGAGTCTTCTAACC
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SSIV 54/10 M        GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 132/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 45/10 M        GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 270/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 237/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 294/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 247/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 18/10 M        GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 206/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 226/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 25/06 M        GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 54/04 M        GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
SSIV 157/09 M       GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
Haseluenne 2003     GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCGGGCCCCCTCAAAGC
AMexico 2009        GAGGTCGAAACGTACGTTCTTTCTATCATCCCGTCAGGCCCCCTCAAAGC
                    ********************************** **************

SSIV 54/10 M        CGAGATCGCGCAGAGACTGGAGGGTGTTTTTTGCGGGGAAGAACACAGAT
SSIV 132/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GTGGGGAAGAACATGGAT
SSIV 45/10 M        CGAGATCGCGCAGAGACTGGAAGGTGTTTTT-GCGGGGAAGAACACAGAT
SSIV 270/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAAAACACAGAT
SSIV 237/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAAAACACAGAT
SSIV 294/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTATTT-GCAGGGAAAAACACAGAT
SSIV 247/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAAAACACAGAT
SSIV 18/10 M        CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAAAACACAGAT
SSIV 206/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAAAACACAGAT
SSIV 226/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAAAACACAGAT
SSIV 25/06 M        CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAGAACACAGAT
SSIV 54/04 M        CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAGAACACAGAT
SSIV 157/09 M       CGAGATCGCGCAGAGACTGGAGGGTGTTTTT-GCAGGGAAAAACACAGAT
Haseluenne 2003     CGAGATCGCGCAGAGACTGGAAGGTGTTTTT-GCAGGGAAGAACACAGAT
AMexico 2009        CGAGATCGCGCAGAGACTGGAAAGTGTCTTT-GCAGGAAAGAACACAGAT
                    **********************  **** *** *  ** ** ****  ***

SSIV 54/10 M        CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 132/09 M       CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 45/10 M        CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 270/09 M       CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 237/09 M       CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 294/09 M       CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 247/09 M       CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 18/10 M        CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 206/09 M       CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 226/09 M       CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 25/06 M        CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 54/04 M        CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCGATCCTGTCACCTCT
SSIV 157/09 M       CTTGAGGCTCTCATGGAATGGTTAAAGACAAGACCGATCCTGTCACCTCT
Haseluenne 2003     CTTGAGGCTCTCATGGAGTGGCTAAAGACAAGACCAATCCTGTCACCTCT
AMexico 2009        CTTGAGGCTCTCATGGAATGGCTAAAGACAAGACCAATCTTGTCACCTCT
                    ***************** *** ************* *** **********

SSIV 54/10 M        GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 132/09 M       GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 45/10 M        GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 270/09 M       GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 237/09 M       GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 294/09 M       GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 247/09 M       GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 18/10 M        GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 206/09 M       GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 226/09 M       GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 25/06 M        GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 54/04 M        GACTAAGGGAATTCTGGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
SSIV 157/09 M       GACTAAGGGAATTCTAGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
Haseluenne 2003     GACTAAGGGAATTCTGGGATTCGTGTTCACGCTCACCGTGCCCAGTGAGC
AMexico 2009        GACTAAGGGAATTTTAGGATTTGTGTTCACGCTCACCGTGCCCAGTGAGC
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SSIV 54/10 M        GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 132/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 45/10 M        GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 270/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 237/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAAATGCCCTAAACGGAAATGG
SSIV 294/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 247/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 18/10 M        GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 206/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 226/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 25/06 M        GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAACGG
SSIV 54/04 M        GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
SSIV 157/09 M       GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAACGGAAATGG
Haseluenne 2003     GAGGACTGCAGCGTAGACGCTTTGTTCAAAA-TGCCCTAAATGGAAATGG
AMexico 2009        GAGGACTGCAGCGTAGACGCTTTGTCCAAAA-TGCCCTAAATGGGAATGG
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SSIV 54/10 M        GGACCCTAATAACATGGATAGAGCAGTTAAATTATACAAGAAACTAAAAA
SSIV 132/09 M       GGACCCTAATAACATGGATAGAGCAGTTAAATTATACAAGAAGCTAAAAA
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SSIV 45/10 M        GGACCCTAATAACATGGATAGAGCAGTTAAATTATACAAGAAGCTAAAAA
SSIV 270/09 M       GGACCCTAATAACATGGATAGAGCAGTCAAATTATATAAGAAGATAAAAA
SSIV 237/09 M       GGACCCTAATAACATGGATAGAGCAGTCAAATTATATAAGAAGATAAAAA
SSIV 294/09 M       GGACCCTAATAACATGGATAGAGCAGTCAAATTATACAAGAAGCTAAAAA
SSIV 247/09 M       GGACCCTAATAACATGGATAGAGCAGCCAAATTATACAAGAAGCTAAAAA
SSIV 18/10 M        GGACCCTAATAACATGGATAGAGCAGTCAAATTATACAAGAAGCTAAAAA
SSIV 206/09 M       GGACCCTAATAACATGGATAGAGCAGTCAAATTATACAAGAAGCTAAAAA
SSIV 226/09 M       GGACCCTAATAACATGGATAGAGCAGTCAAATTATACAAGAAGCTAAAAA
SSIV 25/06 M        GGACCCTAATAACATGGATAGAGCAGTCAAATTATACAAGAAGCTAAAAA
SSIV 54/04 M        GGACCCTAATAACATGGATAGAGCAGTCAAATTATACAAGAAGCTAAAAA
SSIV 157/09 M       GGACCCTAACAACATGGATAGAGCAGTCAAATTATACAAGAAGCTAAAAA
Haseluenne 2003     GGACCCGAATAACATGGATAGAGCAGTCAAATTATACAAGAAGCTCAAGA
AMexico 2009        GGACCCGAACAACATGGATAGAGCAGTTAAACTATACAAGAAGCTCAAAA
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SSIV 54/10 M        GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 132/09 M       GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 45/10 M        GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 270/09 M       GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 237/09 M       GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 294/09 M       GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 247/09 M       GAGAAATAACATTCCATGGGGCCAAGGAAGTATCGCTAAGCTACTCAACT
SSIV 18/10 M        GAGAAATAACATTCCATGGGGCCAAGGAAGTATCGCTAAGCTACTCAACT
SSIV 206/09 M       GAGAAATAACMTTCCATGGGGCCAAGGAAGTATCRCTAAGCTACTCAACT
SSIV 226/09 M       GAGAAATAACCTTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 25/06 M        GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 54/04 M        GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
SSIV 157/09 M       GAGAAATAACATTCCATGGGGCCAAGGAAGTATCACTAAGCTACTCAACT
Haseluenne 2003     GGGAAATAACGTTCCATGGGGCCAAGGAGGTGTCACTAAGCTACTCAACT
AMexico 2009        GAGAAATAACGTTCCATGGGGCCAAGGAGGTGTCACTAAGCTATTCAACT
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SSIV 54/10 M        GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 132/09 M       GGTGCCCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 45/10 M        GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 270/09 M       GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 237/09 M       GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 294/09 M       GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 247/09 M       GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 18/10 M        GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 206/09 M       GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 226/09 M       GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 25/06 M        GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 54/04 M        GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
SSIV 157/09 M       GGTGCTCTTGCCAGTTGCATGGGTCTCATATACAATAGAATGGGAACAGT
Haseluenne 2003     GGTGCACTTGCCAGTTGCATGGGCCTCATATACAACAGAATGGGAACAGT
AMexico 2009        GGTGCACTTGCCAGTTGCATGGGCCTCATATACAACAGGATGGGAACAGT
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SSIV 54/10 M        GACCACAGAAGCTGCCTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 132/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 45/10 M        GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 270/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 237/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 294/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 247/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 18/10 M        GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 206/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAAYTTGTGAGCAGATCG
SSIV 226/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAATTTGTGAGCAGATCG
SSIV 25/06 M        GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 54/04 M        GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
SSIV 157/09 M       GACCACAGAAGCTGCTTTTGGCCTAGTGTGTGCAACTTGTGAGCAGATCG
Haseluenne 2003     GACCACAGAGGCTGCTTTTGGCCTAGTGTGTGCCACTTGTGAGCAGATCG
AMexico 2009        GACCACAGAAGCTGCTTTTGGTCTAGTGTGTGCCACTTGTGAACAGATTG
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SSIV 54/10 M        CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAATCCA
SSIV 132/09 M       CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAATCCA
SSIV 45/10 M        CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 270/09 M       CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 237/09 M       CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 294/09 M       CTGATTCACAGCATCGATCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 247/09 M       CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 18/10 M        CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 206/09 M       CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 226/09 M       CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 25/06 M        CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 54/04 M        CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACCACCAACCCA
SSIV 157/09 M       CTGATTCACAGCATCGGTCACACAGACAAATGGCAACTACCACCAACCCA
Haseluenne 2003     CTGATTCACAGCATCGGTCACACAGACAAATGGCTACTACTACCAACCCA
AMexico 2009        CTGATTCACAGCATCGGTCTCACAGACAGATGGCTACTACCACCAATCCA
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SSIV 54/10 M        CTAATTAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 132/09 M       CTAATTAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 45/10 M        CTAATTAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 270/09 M       CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 237/09 M       CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 294/09 M       CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
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SSIV 247/09 M       CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 18/10 M        CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 206/09 M       CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 226/09 M       CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 25/06 M        CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 54/04 M        CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
SSIV 157/09 M       CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACTGCTAAAGC
Haseluenne 2003     CTAATCAGGCATGAAAACAGAATGGTACTGGCTAGCACTACAGCTAAGGC
AMexico 2009        CTAATCAGGCATGAAAACAGAATGGTGCTGGCTAGCACTACGGCAAAGGC
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SSIV 54/10 M        TATGGAACAGATGGCTGGGTCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 132/09 M       TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 45/10 M        TATGGAACAGATGGCTGGGTCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 270/09 M       TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 237/09 M       TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 294/09 M       TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 247/09 M       TATGGAACAGATGGCTGGATCAAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 18/10 M        TATGGAACAGATGGCTGGATCAAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 206/09 M       TATGGAACAGATGGCTGGATCRAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 226/09 M       TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 25/06 M        TATGGAACAGATGGCTGGATCTAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 54/04 M        TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
SSIV 157/09 M       TATGGAACAGATGGCAGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
Haseluenne 2003     TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCAGAGGCCATGGAGG
AMexico 2009        TATGGAACAGATGGCTGGATCGAGTGAACAGGCAGCGGAGGCCATGGAGG
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SSIV 54/10 M        TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 132/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 45/10 M        TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 270/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 237/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 294/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 247/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 18/10 M        TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 206/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 226/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 25/06 M        TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 54/04 M        TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
SSIV 157/09 M       TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGAACA
Haseluenne 2003     TTGCCAGTCAGACTAGGCAGATGGTGCATGCAATGAGAACAATTGGGACA
AMexico 2009        TTGCTAATCAGACTAGGCAGATGGTACATGCAATGAGAACTATTGGGACT
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SSIV 54/10 M        CATCCCAGCTCCAGTGCCGGTTTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 132/09 M       CATCCCAGCTCCAGTGCCGGTTTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 45/10 M        CATCCCAGCTCCAGTGCCGGTTTGAAAGATGACCTTCTTGAAAATTTACA
SSIV 270/09 M       CATCCCAGCTCCAGTACCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 237/09 M       CATCCCAGCTCCAGTACCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 294/09 M       CATCCCAGCTCCAGTACCGGTCTGAAAGACGACCTTCTTGAAAATTTGCA
SSIV 247/09 M       CATCCCAGCTCCAGTACCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 18/10 M        CATCCCAGCTCCAGTACCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 206/09 M       CATCCCAGCTCCAGTRCCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 226/09 M       CATCCCAGCTCCAGTGCCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 25/06 M        CATCCCAGCTCCAGTGCCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 54/04 M        CATCCCAGCTCCAGTGCCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
SSIV 157/09 M       CATCCCAGCTCCAGTGCCGGTCTGAAAGATGACCTTCTTGAAAATTTACA
Haseluenne 2003     CATCCTAGCTCCAGTGCCGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
AMexico 2009        CATCCTAGCTCCAGTGCTGGTCTGAAAGATGACCTTCTTGAAAATTTGCA
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SSIV 54/10 M        GGCCTACCAGAAACGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 132/09 M       GGCCTACCAGAAACGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 45/10 M        GGCCTACCAGAAACGGATGAGAGTGCAAATACAGCGGTTCAAGTGATGCT
SSIV 270/09 M       GGCCTACCAGAAGCGAATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 237/09 M       GGCCTACCAGAAGCGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 294/09 M       GGCCTACCAGAAGCGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 247/09 M       GGCCTACCAGAAGCGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 18/10 M        GGCCTACCAGAAGCGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 206/09 M       GGCCTACCAGAARCGGAYGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 226/09 M       GGCCTACCAGAAACGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 25/06 M        GGCCTACCAGAAACGGATGGGTGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 54/04 M        GGCCTACCAGAAACGGATGGGAGTGCAAATACAGCGGTTCAAGTGATGTT
SSIV 157/09 M       GGCCTACCAGAAACGGATGGGAGTGCAAATACAGCGGTTCCAGTGATGTT
Haseluenne 2003     GGCATACCAGAAACGGATGGGAGTGCAAATACAGCGGTTCAAGTGATCCT
AMexico 2009        GGCCTACCAGAAGCGAATGGGAGTGCAGATGCAGCGATTCAAGTGATCCT
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SSIV 54/10 M        CTCGCCAATGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 132/09 M       ATCGCCAATGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 45/10 M        ATCGCCAATGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 270/09 M       ATCGCCACTGCAGCAAATATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 237/09 M       ATCGCCACTGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 294/09 M       ATCGCCACTGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 247/09 M       ATCGCCACTACAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 18/10 M        ATCGCCACTACAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 206/09 M       ATCGCCACTGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 226/09 M       ATCGCCACTGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
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SSIV 25/06 M        ATCGCCACTGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 54/04 M        ATCGCCACTGCAGCAAACATCATTGGAATCTTGCACCTGATATTGTGGAT
SSIV 157/09 M       ATAGCAACTGCAGCAAACATCATTGGAATCTTGCACCTGATATCGTGGAT
Haseluenne 2003     CTCGCCATTGCAGCGAATATCGTTGGAATCTTGCACCTGATATTGTGGAT
AMexico 2009        CTCGTCATTGCAGCAAATATCATTGGGATCTTGCACCTGATATTGTGGAT
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SSIV 54/10 M        TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 132/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 45/10 M        TCTTGATCGTCTCTTTTTCAAATGTATCTATCGTCGCCTTAAAATACGG-
SSIV 270/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 237/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 294/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 247/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 18/10 M        TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 206/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACNGG
SSIV 226/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACNGG
SSIV 25/06 M        TCTTGATCGTCTTTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 54/04 M        TCTTGATCGTCTTTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
SSIV 157/09 M       TCTTGATCGTCTCTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
Haseluenne 2003     TCTTGATCGTCTTTTTTTCAAATGTATTTATCGTCGCCTTAAA-TACGG-
AMexico 2009        TACTGATCGTCTTTTTTTCAAATGTATTTATCGTCGCTTTAAA-TACGG-
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SSIV 54/10 M        TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 132/09 M       TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 45/10 M        TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTACCTGAGTCCATGAGGGAA
SSIV 270/09 M       TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 237/09 M       TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 294/09 M       TTTGAAAAGAGGACCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 247/09 M       TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAG
SSIV 18/10 M        TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAG
SSIV 206/09 M       TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAG
SSIV 226/09 M       TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 25/06 M        TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 54/04 M        TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
SSIV 157/09 M       TTTGAAAAGAGGGCCTNTCTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
Haseluenne 2003     TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
AMexico 2009        TTTGAAAAGAGGGCCTT-CTACGGAAGGAGTGCCTGAGTCCATGAGGGAA
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SSIV 54/10 M        GAATATCAACAGGAACAGCAGAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 132/09 M       GAATATCAACAGGAACAGCAGAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 45/10 M        GAATATCAACAGGAACAGCAGAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 270/09 M       GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 237/09 M       GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 294/09 M       GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 247/09 M       GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 18/10 M        GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 206/09 M       GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 226/09 M       GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 25/06 M        GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 54/04 M        GAATATCAACAGGAACAGCAAAGTGCTGTGGATGTTGACGATGGTCATTT
SSIV 157/09 M       GAATATCAACAGGAACAGCAGAGTGCTGTGGATGTTGACGATGGTCATTT
Haseluenne 2003     GAATATCAACAGGAACAGCAGAGTGCTGTGGATGTTGACGGTGGTCATTT
AMexico 2009        GAATATCAACAGGAACAGCAGAGTGCTGTGGATGTTGACGATGGTCATTT
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SSIV 54/10 M        TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 132/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 45/10 M        TGTCAACATAGAGCTGGAGTAAANNACTACCTTGTTTCTA-CTGATCACG
SSIV 270/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 237/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 294/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTNNNNACNGATCACG
SSIV 247/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 18/10 M        TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CK-ATCACG
SSIV 206/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 226/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 25/06 M        TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 54/04 M        TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
SSIV 157/09 M       TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CTGATCACG
Haseluenne 2003     TGTCAACATAGAGCTGGAGTAAAAAACTACCTTGTTTCTA-CT-------
AMexico 2009        TGTCAACATAGAGCTAGAGTAA--------------------------
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SSIV 54/10 M        CGT
SSIV 132/09 M       CGT
SSIV 45/10 M        CGT
SSIV 270/09 M       CGT
SSIV 237/09 M       CGT
SSIV 294/09 M       CGT
SSIV 247/09 M       CGT
SSIV 18/10 M        CGT
SSIV 206/09 M       CGT
SSIV 226/09 M       CGT
SSIV 25/06 M        CGT
SSIV 54/04 M        CGT
SSIV 157/09 M       CGT
Haseluenne 2003---
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AMexico 2009---

Legend:
Nucleotide sequences of segment 7. Highlighted in red: sequences affecting amantadine resistance (corresponding to 30A and 34G in the sequence of the translated protein).

Appendix 2b

M1 protein

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SSIV 206/09 M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 226/09 M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 157/09 M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 45/10  M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
Mexico 2009         MSLLTEVETYVLSIIPSGPLKAEIAQRLESVFAGKNTDLEALMEWLKTRP
SSIV 132/09 M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFVGKNMDLEALMEWLKTRP
Haseluenne 2003     MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 25/06 M1       MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 54/04 M1       MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 54/10 M1       MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 294/09 M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 18/10  M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 247/09 M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 270/09 M1      MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
SSIV 237/09 M1_     MSLLTEVETYVLSIIPSGPLKAEIAQRLEGVFAGKNTDLEALMEWLKTRP
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SSIV 206/09 M1      ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 226/09 M1      ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 157/09 M1      ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 45/10 M1       ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
Mexico 2009         ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 132/09 M1      ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
Haseluenne 2003     ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 25/06 M1       ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 54/04 M1       ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 54/10 M1       ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 294/09 M1      ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 18/10 M1       ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 247/09 M1      ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAAKLY
SSIV 270/09 M1      ILSPLTKGILGFVFTLTVPSERGLQRRRFVQNALNGNGDPNNMDRAVKLY
SSIV 237/09 M1_     ILSPLTKGILGFVFTLTVPSERGLQRRRFVQKCPKRKWGPYNMDRAVKLY
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SSIV 206/09 M1      KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAX
SSIV 226/09 M1      KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAI
SSIV 157/09 M1      KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 45/10 M1       KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
Mexico 2009         KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 132/09 M1      KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
Haseluenne 2003     KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 25/06 M1       KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 54/04 M1       KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 54/10 M1       KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 294/09 M1      KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 18/10 M1       KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 247/09 M1      KKLKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 270/09 M1      KKIKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
SSIV 237/09 M1_     KKIKREITFHGAKEVSLSYSTGALASCMGLIYNRMGTVTTEAAFGLVCAT
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SSIV 206/09 M1      CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 226/09 M1      CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 157/09 M1      CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 45/10 M1       CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
Mexico 2009         CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 132/09 M1      CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
Haseluenne 2003     CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 25/06 M1       CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 54/04 M1       CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 54/10 M1       CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 294/09 M1      CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 18/10 M1       CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 247/09 M1      CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 270/09 M1      CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
SSIV 237/09 M1_     CEQIADSQHRSHRQMATTTNPLIRHENRMVLASTTAKAMEQMAGSSEQAA
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SSIV 206/09 M1      EAMEVASQTRQMVHAMRTIGTHPSSSXGLKDDLLENLQAYQKRXGVQIQR
SSIV 226/09 M1      EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQIQR
SSIV 157/09 M1      EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQIQR
SSIV 45/10 M1       EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMRVQIQR
Mexico 2009         EAMEVANQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQMQR
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SSIV 132/09 M1      EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQIQR
Haseluenne 2003     EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQIQR
SSIV 25/06 M1       EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQIQR
SSIV 54/04 M1       EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQIQR
SSIV 54/10 M1       EAMEVASQTRQMVHAMRTIGTHPSSSAGLKDDLLENLQAYQKRMGVQIQR
SSIV 294/09 M1      EAMEVASQTRQMVHAMRTIGTHPSSSTGLKDDLLENLQAYQKRMGVQIQR
SSIV 18/10 M1       EAMEVASQTRQMVHAMRTIGTHPSSSTGLKDDLLENLQAYQKRMGVQIQR
SSIV 247/09 M1      EAMEVASQTRQMVHAMRTIGTHPSSSTGLKDDLLENLQAYQKRMGVQIQR
SSIV 270/09 M1      EAMEVASQTRQMVHAMRTIGTHPSSSTGLKDDLLENLQAYQKRMGVQIQR
SSIV 237/09 M1_     EAMEVASQTRQMVHAMRTIGTHPSSSTGLKDDLLENLQAYQKRMGVQIQR
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SSIV 206/09 M1      FK
SSIV 226/09 M1      FK
SSIV 157/09 M1      FQ
SSIV 45/10 M1       FK
Mexico 2009         FK
SSIV 132/09 M1      FK
Haseluenne 2003     FK
SSIV 25/06 M1       FK
SSIV 54/04 M1       FK
SSIV 54/10 M1       FK
SSIV 294/09 M1      FK
SSIV 18/10 M1       FK
SSIV 247/09 M1      FK
SSIV 270/09 M1      FK
SSIV 237/09 M1_     FK
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Legend:
Amino acid sequences of M1.

Appendix 2c

M2 protein

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SSIV 237/09 M2      MSLLTEVETPTRSGWECKYSGSSDVIATAANIIGILHLILWILDRLFFKC
SSIV 294/09 M2      MSLLTEVETPTRSGWECKYSGSSDVIATAANIIGILHLILWILDRLFFKC
SSIV 206/09 M2      MSLLTEVETPTRSGWECKYSGSSDVIATAANIIGILHLILWILDRLFFKC
SSIV 18/10 M2       MSLLTEVETPTRSGWECKYSGSSDVIATTANIIGILHLILWILDRLFFKC
SSIV 247/09 M2      MSLLTEVETPTRSGWECKYSGSSDVIATTANIIGILHLILWILDRLFFKC
SSIV 270/09 M2      MSLLTEVETPTRSEWECKYSGSSDVIATAANIIGILHLILWILDRLFFKC
SSIV 226/09 M2      MSLLTEVETPTRNGWECKYSGSSDVIATAANIIGILHLILWILDRLFFKC
SSIV 54/04 M2       MSLLTEVETPTRNGWECKYSGSSDVIATAANIIGILHLILWILDRLFFKC
SSIV 25/06 M2       MSLLTEVETPTRNGWVCKYSGSSDVIATAANIIGILHLILWILDRLFFKC
SSIV 157/09 M2      MSLLTEVETPTRNGWECKYSGSSDVIATAANIIGILHLISWILDRLFFKC
SSIV 132/09 M2      MSLLTEVETPTRNGWECKYSGSSDVIANAANIIGILHLILWILDRLFFKC
SSIV 45/10 M2       MSLLTEVETPTRNGWECKYSGSSDAIANAANIIGILHLILWILDRLFFKC
SSIV 54/10 M2       MSLLTEVETPTRNGWECKYSGSSDVLANAANIIGILHLILWILDRLFFKC
Haseluenne 2003     MSLLTEVETHTRNGWECKYSGSSDPLAIAANIVGILHLILWILDRLFFKC
Mexico 2009         MSLLTEVETPTRSEWECRCSDSSDPLVIAANIIGILHLILWITDRLFFKC
                    ********* **.   *: *.*** :. :***:****** ** *******

SSIV 237/09 M2      IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 294/09 M2      IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 206/09 M2      IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 18/10 M2       IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 247/09 M2      IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 270/09 M2      IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 226/09 M2      IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 54/04 M2       IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 25/06 M2       IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 157/09 M2      IYRRLKYGLKRGPSTEGVPVSMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 132/09 M2      IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 45/10 M2       IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
SSIV 54/10 M2       IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
Haseluenne 2003     IYRRLKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDGGHFVNIELE
Mexico 2009         IYRRFKYGLKRGPSTEGVPESMREEYQQEQQSAVDVDDGHFVNIELE
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Legend:
Amino acid sequences of the M2 protein. 30A and 34G are highlighted in red.

Appendix 3a

Segment 6

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SSIV 54/10 NA     ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
SSIV 132/09 NA    ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
SSIV 199/09 NA    ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
SSIV 247/09 NA    ACNCGTGATCAGNANAAGCANGAGTTCAAAATGAATCCNAATCAGAANAT
SSIV 45/10 NA     ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
SSIV 18/10 NA     ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCNAATCAGAAGAT
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SSIV 206/09 NA    ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
SSIV 270/09 NA    ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
SSIV 294/09 NA    ACGCGTGATCAGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
Haseluenne 2003   ----------AGCAAAAGCAGGAGTTCAAAATGAATCCAAATCAGAAGAT
Mexico 2009       ----------------------------AAATGAATCCAAACCAAAAGAT
                                              ********** ** ** ** **

SSIV 54/10 NA     AATAACCATTAGTTCGATCTGTTTGATAAATGGAATTACTAGCTTGATAT
SSIV 132/09 NA    AATAATCATCAGTTCGATCTGTTTGATAAATGGAATTACTAGCTTGATAT
SSIV 199/09 NA    AATAATCATTAGTTCGATCTGTTTGATAAATGGAATTACTAGCTTGATAT
SSIV 247/09 NA    AATAATCATTANTTCGATCTGTTTGATAAATGGAATTACTAGCTTGATAT
SSIV 45/10 NA     AATAATCATTAGTTCGATCTGTTTGATAAATGGAATTACTAGCTTGATAT
SSIV 18/10 NA     AATAATCATTAGTTCGATCTGTTTGATAAATGGAATTGCTAGCTTGATAT
SSIV 206/09 NA    AATAATCATTAGTTCGATCTGTTTGATAAATGGAATTGCTAGCTTGATAT
SSIV 270/09 NA    AATAATCATTAGTTCGATCTGTTTGATAAATGGAATTGCTGGCTTGATAT
SSIV 294/09 NA    AATAATCATTAGTTCGATCTGTTTGATAAGTGGAATTGCTAGCTTGATAT
Haseluenne 2003   AATAATCATTAGTTCGATCTGTATGATAAATGGAATTGCTAGCTTGATAT
Mexico 2009       AATAACCATTGGTTCGGTCTGTATGACAATTGGAATGGCTAACTTAATAT
                  ***** ***   **** ***** *** ** ******  **  *** ****

SSIV 54/10 NA     TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
SSIV 132/09 NA    TACAAATTGGGAACATAATCTCAATATGGATTAGTCATTCAATTCAAATT
SSIV 199/09 NA    TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
SSIV 247/09 NA    TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCNATTCAAATT
SSIV 45/10 NA     TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
SSIV 18/10 NA     TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
SSIV 206/09 NA    TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
SSIV 270/09 NA    TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
SSIV 294/09 NA    TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
Haseluenne 2003   TACAAATTGGGAACATAATCTCAATATGGATTAGCCATTCAATTCAAATT
Mexico 2009       TACAAATTGGAAACATAATCTCAATATGGATTAGCCACTCAATTCAACTT
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SSIV 54/10 NA     GGGAACCAAAACCAGACTGAAACATGCAGCCAAAACATCATTACTTATGA
SSIV 132/09 NA    GGGAACCAAAACCAGACTGAAACATGCAGCCAAAACATCATTACTTATGA
SSIV 199/09 NA    GGGAACCAAAACCAGACTGAAACATGCAGCCAAAACATCATTACTTATGA
SSIV 247/09 NA    GGNAACCAAAACCAGACTGAAACATGCAGCCAAAACATCATTACTTATGA
SSIV 45/10 NA     GGGAACCAAAACCAGACTGAAACATGCAGCCAAAACATCATTACTTATGA
SSIV 18/10 NA     GGGAACCAAAACCAGACTGAAACATGCAGCCAGAACATCATTACTTATGA
SSIV 206/09 NA    GGGAACCAAAACCAGACTGAAACATGCAGCCAGAACATCATTACTTATGA
SSIV 270/09 NA    GGGAACCAAAACCAGACTGAAACATGCAGCCAGAACATCATTACTTACGA
SSIV 294/09 NA    GGGAACCAAAACCAGACTGAAACATGCAGCCAGAACATCATTACTTACGA
Haseluenne 2003   GGGAACCAAAACCAGACTGAAACATGCAATCAAAGCGTCATTACTTACGA
Mexico 2009       GGGAACCAAAATCAGATTGAAACATGCAATCAAAGCGTCATTACTTATGA
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SSIV 54/10 NA     AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
SSIV 132/09 NA    AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
SSIV 199/09 NA    AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
SSIV 247/09 NA    AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
SSIV 45/10 NA     AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
SSIV 18/10 NA     AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
SSIV 206/09 NA    AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
SSIV 270/09 NA    AAACAACACTTGGGTAAATCATACATATGTTAACATCAGCAACAACAATT
SSIV 294/09 NA    AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
Haseluenne 2003   AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACAACAATT
Mexico 2009       AAACAACACTTGGGTAAATCAGACATATGTTAACATCAGCAACACCAACT
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SSIV 54/10 NA     TTGTTGCTGAACAGGCAGTTGTCTCANTGAAGTTAGCGGGCAGTTCGTCT
SSIV 132/09 NA    TTGTTGCTGAACAGGCAGTTGTCTCAATGAAGTTAGCGGGCAGTTCATCT
SSIV 199/09 NA    TTGTTGCTGAACAGGCAGTTGTCTCAATGAAGTTAGCGGGCAGTTCGCCT
SSIV 247/09 NA    TTGTTGCTGANCAGGCAGTTGTCTCAATGAAGTTAGCGGGCNGTTCGCCT
SSIV 45/10 NA     TTGTTGCTGAACAAGCTGTTGTCTCAATGAAGTTAGCGGGCAGTTCGTCT
SSIV 18/10 NA     TTGTTGCTGAACAGGCAGTTGTCTCAATAAAGTTAGCAGGCAGTTCGTCC
SSIV 206/09 NA    TTGTTGCTGAACAGGCAGTTGTCTCAATAAAGTTAGCAGGCAGTTCGTCC
SSIV 270/09 NA    TTGTTGCTGAACAGGCAGTTGTCTCAACAAAGTTAGCGGGCAGTTCGTCC
SSIV 294/09 NA    TTGTTGCTGAACAGGCAGTTGTCCCAATAAAGTTAGCGGGCAGTTCGTCC
Haseluenne 2003   TTGTTGCTGAACAGGCAGTGGTTTCAGTGAAGTTAGCGGGCAGTTCTCCT
Mexico 2009       TTGCTGCTGGACAGTCAGTGGTTTCCGTGAAATTAGCGGGCAATTCCTCT
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SSIV 54/10 NA     CTCTGCCCCGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 132/09 NA    CTCTGCCCCGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 199/09 NA    CTCTGCCCCGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 247/09 NA    CTCTGCCCCGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 45/10 NA     CTCTGCCCCGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 18/10 NA     CTCTGCCCTGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 206/09 NA    CTCTGCCCTGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 270/09 NA    CTCTGCCCTGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
SSIV 294/09 NA    CTCTGCCCTGTTAGTGGGTGGGCTATATACAGTAAAGATAACAGTGTAAG
Haseluenne 2003   CTCTGCTCTGTTAGTGGGTGGGCCATATACAGTAAAGATAACAGTGTAAG
Mexico 2009       CTCTGCCCTGTTAGTGGATGGGCTATATACAGTAAAGACAACAGTGTAAG
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SSIV 54/10 NA     AATCGGTTCCAAGGGAGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
SSIV 132/09 NA    AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
SSIV 199/09 NA    AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
SSIV 247/09 NA    AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
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SSIV 45/10 NA     AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
SSIV 18/10 NA     AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAGCCATTCATATCAT
SSIV 206/09 NA    AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
SSIV 270/09 NA    AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
SSIV 294/09 NA    AGTCGGTTCCAAGGGGGATGTGTTTGTCATAAGAGAACCATTCATCTCAT
Haseluenne 2003   AATCGGTTCCAGGGGGGATGTGTTTGTCATAAGAGAGCCATTCATCTCAT
Mexico 2009       AATCGGTTCCAAGGGGGATGTGTTTGTCATAAGGGAACCATTCATATCAT
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SSIV 54/10 NA     GCTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
SSIV 132/09 NA    GCTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
SSIV 199/09 NA    GCTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
SSIV 247/09 NA    GCTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCANGGGGCCCTGCTG
SSIV 45/10 NA     GCTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
SSIV 18/10 NA     GTTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
SSIV 206/09 NA    GTTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
SSIV 270/09 NA    GTTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
SSIV 294/09 NA    GTTCCCACTTGGAATGTAGAACCTTCTTCTTGACTCAAGGGGCCCTGCTG
Haseluenne 2003   GCTCCCACTTGGAATGTAGAACTTTCTTCTTGACTCAAGGGGCCCTGCTG
Mexico 2009       GCTCCCCCTTGGAATGCAGAACCTTCTTCTTGACTCAAGGGGCCTTGCTA
                  * **** ********* ***** ************** ****** ****

SSIV 54/10 NA     AATGACAAACACTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
SSIV 132/09 NA    AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
SSIV 199/09 NA    AATGACAAACATTCTAATGGAACTATTAAAGACAGAAGCCCCTACCGAAC
SSIV 247/09 NA    AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
SSIV 45/10 NA     AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
SSIV 18/10 NA     AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
SSIV 206/09 NA    AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
SSIV 270/09 NA    AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
SSIV 294/09 NA    AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTACCGAAC
Haseluenne 2003   AATGACAAACATTCTAATGGAACCATTAAAGACAGAAGCCCCTATCGAAC
Mexico 2009       AATGACAAACATTCCAATGGAACCATTAAAGACAGGAGCCCATATCGAAC
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SSIV 54/10 NA     CCTGATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 132/09 NA    CCTGATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 199/09 NA    CCTGATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 247/09 NA    CCTGNTGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 45/10 NA     CCTGATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 18/10 NA     CCTTATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 206/09 NA    CCTTATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 270/09 NA    CCTTATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
SSIV 294/09 NA    CCTTATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCATACAACTCAAGAT
Haseluenne 2003   CCTGATGAGCTGTCCTATTGGTGAAGTCCCCTCTCCGTACAACTCAAGAT
Mexico 2009       CCTAATGAGCTGTCCTATTGGTGAAGTTCCCTCTCCATACAACTCAAGAT
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SSIV 54/10 NA     TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGAACCAGTTGG
SSIV 132/09 NA    TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGAACCAGTTGG
SSIV 199/09 NA    TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATAATGGAACCAGTTGG
SSIV 247/09 NA    TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGAACCAGTTGG
SSIV 45/10 NA     TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGAACCAGTTGG
SSIV 18/10 NA     TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGCACCAGTTGG
SSIV 206/09 NA    TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGCACCAGTTGG
SSIV 270/09 NA    TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGCACCAGTTGG
SSIV 294/09 NA    TTGAGTCAGTTGCTTGGTCAGCAAGTGCTTGCCATGATGGCACCAGTTGG
Haseluenne 2003   TTGAGTCAGTTGCTTGGTCAGCAAGCGCTTGCCATGATGGCACCAGTTGG
Mexico 2009       TTGAGTCAGTCGCTTGGTCAGCAAGTGCTTGTCATGATGGCATCAATTGG
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SSIV 54/10 NA     TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 132/09 NA    TTGACAATTGGAATTTCTGGGCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 199/09 NA    TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 247/09 NA    TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 45/10 NA     TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 18/10 NA     TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 206/09 NA    TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 270/09 NA    TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
SSIV 294/09 NA    TTGACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTATTGAA
Haseluenne 2003   TTGACAATTGGAATTTCTGGCCCAGATAATGGGGCAGTGGCTGTATTGAA
Mexico 2009       CTAACAATTGGAATTTCTGGCCCAGACAATGGGGCAGTGGCTGTGTTAAA
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SSIV 54/10 NA     ATACAATGACATAATCACAGACACTATCAAGAGTTGGAGAAACAACATAT
SSIV 132/09 NA    ATACAATGACATAATCACAGACACTATCAAGAGTTGGAGAAACAACATAT
SSIV 199/09 NA    ATACAATGACATAATCACAGACACTATCAAGAGTTGGAGAAACAACATAT
SSIV 247/09 NA    ATACAATGACATAATCACAGACACTATCAAGAGTTGGAGAAACAACATAT
SSIV 45/10 NA     ATACAATGACATAATCACAGACACTATCAGGAGTTGGAGAAACAACATAT
SSIV 18/10 NA     ATACAATGACATAATAACAGACACTATCAAGAGTTGGAGAAACAACATAT
SSIV 206/09 NA    ATACAATGACATAATAACAGACACTATCAAGAGTTGGAGAAACAACATAT
SSIV 270/09 NA    ATACAATGACATAATAACAGACACTATCAAGAGTTGGAGAAACAACATAT
SSIV 294/09 NA    ATACAATGACATAATAACAGACACTATCAAGAGTTGGAGAAACAACATAT
Haseluenne 2003   ATACAACGACATAATAACAGACACTATCAAGAGTTGGAGAAACAACATAT
Mexico 2009       GTACAACGGCATAATAACAGACACTATCAAGAGTTGGAGAAACAATATAT
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SSIV 54/10 NA     TGAGAACACAAGAGTCTGAATGTGTATGCGTAAATGGCTCTTGCTTTACT
SSIV 132/09 NA    TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGCTTTACT
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SSIV 199/09 NA    TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGCTTTACT
SSIV 247/09 NA    TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGCTTTACT
SSIV 45/10 NA     TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGCTTCACT
SSIV 18/10 NA     TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGTTATACT
SSIV 206/09 NA    TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGCTTTACT
SSIV 270/09 NA    TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGCTTTACT
SSIV 294/09 NA    TGAGAACACAAGAGTCTGAATGTGCATGCGTAAATGGCTCTTGCTTCACT
Haseluenne 2003   TGAGAACACAAGAGTCTGAATGTGCATGCGTGAATGGCTCTTGCTTTACT
Mexico 2009       TGAGAACACAAGAGTCTGAATGTGCATGTGTAAATGGTTCTTGCTTTACT
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SSIV 54/10 NA     GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCGTACAAGATCTTCAA
SSIV 132/09 NA    GTAATGACCGATGGACCAAATAATGGGCAGGCCTCGTACAAGATCTTCAA
SSIV 199/09 NA    GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCGTACAAAATCTTCAA
SSIV 247/09 NA    GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCGTACAAGATCTTCAA
SSIV 45/10 NA     GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCGTACAAGATCTTCAA
SSIV 18/10 NA     GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCGTACAAGATCTTCAA
SSIV 206/09 NA    GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCGTACAAGATCTTCAA
SSIV 270/09 NA    GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCGTACAAGATCTTCAA
SSIV 294/09 NA    GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCATACAAGATCTTCAA
Haseluenne 2003   GTAATGACCGATGGACCAAGTAATGGGCAGGCCTCATACAAGATCTTCAA
Mexico 2009       GTAATGACCGATGGACCAAGTAATGGACAGGCCTCATACAAGATCTTCAG
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SSIV 54/10 NA     AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAGTTGAATGCCCCTAATT
SSIV 132/09 NA    AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAGTTGAATGCCCCTAATT
SSIV 199/09 NA    AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAGTTGAATGCCCCTAATT
SSIV 247/09 NA    AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAGTTGAATGCCCCTAATT
SSIV 45/10 NA     AATAGAAAAAGGGAAAGTAGTCAAATCAGTCGAGTTGAATGCCCCTAATT
SSIV 18/10 NA     AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAATTGAATGCCCCTAATT
SSIV 206/09 NA    AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAATTGAATGCCCCTAATT
SSIV 270/09 NA    AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAATTGAATGCCCCTAATT
SSIV 294/09 NA    AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAATTGAATGCCCCTAATT
Haseluenne 2003   AATAGAAAAGGGGAAAGTAGTCAAATCAGTCGAGTTGAATGCCCCTAATT
Mexico 2009       AATAGAAAAGGGAAAGATAGTCAAATCAGTCGAAATGAATGCCCCTAATT
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SSIV 54/10 NA     ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
SSIV 132/09 NA    ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
SSIV 199/09 NA    ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
SSIV 247/09 NA    ATCACTATGAGGAATGTTCCTGTTATCCTGATTNTGGTGAAATCATATGT
SSIV 45/10 NA     ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
SSIV 18/10 NA     ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
SSIV 206/09 NA    ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
SSIV 270/09 NA    ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
SSIV 294/09 NA    ATCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
Haseluenne 2003   ACCACTATGAGGAATGTTCCTGTTATCCTGATTCTGGTGAAATCATATGT
Mexico 2009       ATCACTATGAGGAATGCTCCTGTTATCCTGATTCTAGTGAAATCACATGT
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SSIV 54/10 NA     GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
SSIV 132/09 NA    GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
SSIV 199/09 NA    GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
SSIV 247/09 NA    GTGTGCAGGGACAATTGGCANGGATNGAATCGACCGTGGGTGTCTTTCAA
SSIV 45/10 NA     GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
SSIV 18/10 NA     GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
SSIV 206/09 NA    GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
SSIV 270/09 NA    GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
SSIV 294/09 NA    GTGTGCAGGGACAATTGGCATGGATCGAATCGACCGTGGGTGTCTTTCAA
Haseluenne 2003   GTATGCAGGGACAATTGGCATGGCTCGAATCGACCGTGGGTGTCTTTCAA
Mexico 2009       GTGTGCAGGGATAACTGGCATGGCTCGAATCGACCGTGGGTGTCTTTCAA
                  ** ******** ** ***** ** * ************************

SSIV 54/10 NA     TCAGGATCTGGAGTATCAAATAGGATACATATGCAGTGGAATTCTCGGAG
SSIV 132/09 NA    TCAGAATCTGGAGTATCAAATAGGATACATATGCAGTGGAATTCTCGGAG
SSIV 199/09 NA    TCAGAATCTGGAGTATCAAATAGGATACATATGCAGTGGAATTCTCGGAG
SSIV 247/09 NA    TCAGAATNTGGAGTATCAAATAGGANACATATGCAGTGGAATTCTCGGAG
SSIV 45/10 NA     TCAGAATCTGGAGTATCAAATAGGATACATATGCAGTGGAATTCTCGGAG
SSIV 18/10 NA     TCAGAATCTGGAGTATCAAATAGGATACATATGCAGTGGAGTTCTCGGAG
SSIV 206/09 NA    TCAGAATCTGGAGTATCAAATAGGATACATATGCAGTGGAGTTCTCGGAG
SSIV 270/09 NA    TCAGAATCTGGAGTATCAAATAGGATACATATGCAGTGGAGTTCTCGGAG
SSIV 294/09 NA    TCAGAATCTGGAGTATCAAATAGGATACATATGCAGTGGAGTTCTCGGAG
Haseluenne 2003   TCAGAATCTGGAGTATCAAATAGGATATATATGCAGTGGGGTTCTCGGAG
Mexico 2009       CCAGAATCTGGAATATCAGATAGGATACATATGCAGTGGGATTTTCGGAG
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SSIV 54/10 NA     ATAACCCTCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCGCTT
SSIV 132/09 NA    ATAACCCTCGCCCTAATGATAGAACAGGCAGTTGTGGCCCAGTATCGCTT
SSIV 199/09 NA    ATAACCCTCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCACTT
SSIV 247/09 NA    ATAACCCTCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCACTT
SSIV 45/10 NA     ATAACCCTCGACCTAATGATAGAACAGGCAGTTGTGATCCAGTATCGCTT
SSIV 18/10 NA     ATAACCCGCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCGCTT
SSIV 206/09 NA    ATAACCCGCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCGCTT
SSIV 270/09 NA    ATAACCCGCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCGCTT
SSIV 294/09 NA    ATAACCCGCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCGCTT
Haseluenne 2003   ACAATCCGCGCCCTAATGATAGAACAGGCAGTTGTGGTCCAGTATCGTCT
Mexico 2009       ACAATCCACGCCCTAATGATAAGACAGGCAGTTGTGGTCCAGTATCGTCT
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SSIV 54/10 NA     AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 132/09 NA    AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 199/09 NA    AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 247/09 NA    AATGGAGCTAATGGAGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 45/10 NA     AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 18/10 NA     AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 206/09 NA    AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 270/09 NA    AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
SSIV 294/09 NA    AATGGAGCTAATGGGGTAAAAGGGTTTTCGTTCAAATACGGCAATGGTGT
Haseluenne 2003   AATGGAGCAAATGGGGTAAAAGGGTTTTCGTTTAAATACGGCAATGGTGT
Mexico 2009       AATGGAGCAAATGGAGTAAAAGGATTTTCATTCAAATACGGCAATGGTGT
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SSIV 54/10 NA     TTGGATAGGGAGAACTAAAAGCACTAGTTCAAGGAGCGGTTTTGAGATGA
SSIV 132/09 NA    TTGGATAGGGAGAACTAAAAGCACTAGTTCAAGGAGCGGTTTTGAGATGA
SSIV 199/09 NA    TTGGATAGGGAGAACTAAAAGCACTAGTTCAAGGAGCGGTTTTGAGATGA
SSIV 247/09 NA    TTGGATAGGGAGAANTAAAAGCANTAGTTCAAGGAGCGGTTTTGAGATGA
SSIV 45/10 NA     TTGGATAGGGAGAACTAAAAGCACTAGTTCAAGGAGCGGTTTTGAGATGA
SSIV 18/10 NA     TTGGATAGGGAGAACTAAAAGCACTAGTTCAAGGAGCGGTTTTGAGATGG
SSIV 206/09 NA    TTGGATAGGGAGAACTAAAAGCACTAGTTCAAGGAGCGGTTTTGAGATGA
SSIV 270/09 NA    TTGGATAGGAAGAACTAAAAGCACTAGTTCAAGGAGTGGTTTTGAGATGA
SSIV 294/09 NA    TTGGATAGGGAGAACTAAAAGCACTAGCTCAAGGAGTGGTTTTGAGATGA
Haseluenne 2003   TTGGATAGGGAGAACTAAAAGCACTAGTTCAAGGAGCGGTTTTGAGATGA
Mexico 2009       TTGGATAGGGAGAACTAAAAGCATTAGTTCAAGAAACGGTTTTGAGATGA
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SSIV 54/10 NA     TTTGGGACCCAAACGGATGGACTGGAACAGACAATAACTTCTTAGTAAAG
SSIV 132/09 NA    TTTGGGATCCAAACGGATGGACTGGAACAGACAATAACTTCTTAGTAAAG
SSIV 199/09 NA    TTTGGGATCCAAACGGATGGACTGGAACAGACAATAACTTCTTAGTAAAG
SSIV 247/09 NA    TTTGGGATCCAAACGGATGGACTGGANCAGACAATAACTTCTTAGTAAAG
SSIV 45/10 NA     TTTGGGATCCAAACGGTTGGACTGGAACAGACAATAACTTCTTAGTAAAG
SSIV 18/10 NA     TTTGGGATCCAAACGGATGGACCGGAACAGACAATAACTTCTTAGTAAAG
SSIV 206/09 NA    TTTGGGATCCAAACGGATGGACCGGAACAGACAATAACTTCTTAGTAAAG
SSIV 270/09 NA    TTTGGGATCCAAACGGATGGACCGGAACAGACAATAACTTCTTAGTAAAG
SSIV 294/09 NA    TTTGGGATCCAAACGGATGGACAGGAACAGACAATAACTTCTTAGTAAAG
Haseluenne 2003   TTTGGGACCCAAACGGATGGACTGGAACAGACAATAACTTCTCAGTAAAG
Mexico 2009       TTTGGGATCCGAACGGATGGACTGGGACAGACAATAACTTTTCAATAAAG
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SSIV 54/10 NA     CAAGATATCGTAGGGATGACTGACTGGTCAGGATACAGTGGCAGTTTTGT
SSIV 132/09 NA    CAAGATATCGTAGGGATGACTGACTGGTCAGGATATAGTGGCAGTTTTGT
SSIV 199/09 NA    CAAGATATCGTAGGGATGACTGACTGGTCAGGATACAGTGGCAGTTTTGT
SSIV 247/09 NA    CAAGATATCGTAGGGATGACTGACTGGTCAGGATACAGTGGCAGTTTTGT
SSIV 45/10 NA     CAAGATATCGTAGGAATGACTGATTGGTCAGGATACAGTGGCAGTTTTGT
SSIV 18/10 NA     CAAGATATCGTAGGAATGACTGACTGGTCAGGATACAGTGGCAGTTTTGT
SSIV 206/09 NA    CAAGATATCGTAGGAATGACTGACTGGTCAGGATACAGTGGCAGTTTTGT
SSIV 270/09 NA    CAAGATATCGTAGGAATGACTGACTGGTCAGGATACAGTGGCAGTTTTGT
SSIV 294/09 NA    CAAGATATCGTAGGAATGACTGACTGGTCAGGATACAGTGGCAGTTTTGT
Haseluenne 2003   CAAGATATCGTAGGAATAACTGACTGGTCAGGATACAGCGGGAGTTTTGT
Mexico 2009       CAAGATATCGTAGGAATAAATGAGTGGTCAGGATATAGCGGGAGTTTTGT
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SSIV 54/10 NA     TCAGCATCCAGAACTAACCGGGATGAATTGTATGAGACCTTGCTTCTGGG
SSIV 132/09 NA    TCAGCATCCAGAACTAACCGGGATGAATTGTATGAGACCTTGCTTCTGGG
SSIV 199/09 NA    TCAGCATCCAGAACTAACCGGGATGAATTGTATGAGACCTTGCTTCTGGG
SSIV 247/09 NA    TCAGCATCCAGAACTAACCGGGATGAATNGTATGAGACCTTGNNTNTGGG
SSIV 45/10 NA     TCAGCATCCAGAACTAACCGGGATGAATTGTATAAGACCTTGCTTCTGGG
SSIV 18/10 NA     TCAGCATCCAGAACTAACCGGGATGAATTGTATGAGACCTTGCTTCTGGG
SSIV 206/09 NA    TCAGCATCCAGAACTAACCGGGATGAATTGTATGAGACCTTGCTTCTGGG
SSIV 270/09 NA    TCAGCATCCAGAACTAACCGGGATGAATTGTATGAGACCTTGCTTCTGGG
SSIV 294/09 NA    TCAGCATCCAGAACTAACCGGGATGAATTGTATGAGACCTTGCTTCTGGG
Haseluenne 2003   TCAGCATCCAGAACTAACCGGACTGAATTGTATGAGACCTTGCTTCTGGG
Mexico 2009       TCAGCATCCAGAACTAACAGGGCTGGATTGTATAAGACCTTGCTTCTGGG
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SSIV 54/10 NA     TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
SSIV 132/09 NA    TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
SSIV 199/09 NA    TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
SSIV 247/09 NA    TTGAACTGATCAGAGGGCGACCCAAAGAGNACNCAATCTGGACTAGCGGG
SSIV 45/10 NA     TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
SSIV 18/10 NA     TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
SSIV 206/09 NA    TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
SSIV 270/09 NA    TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
SSIV 294/09 NA    TTGAACTGATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
Haseluenne 2003   TTGAACTAATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
Mexico 2009       TTGAACTAATCAGAGGGCGACCCAAAGAGAACACAATCTGGACTAGCGGG
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SSIV 54/10 NA     AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGAATTGGTCTTG
SSIV 132/09 NA    AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGGATTGGTCTTG
SSIV 199/09 NA    AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGGATTGGTCTTG
SSIV 247/09 NA    AGCAGCATATCCTTTTNTGGTGTAAATAGTGACACTGTGGGTTGGTCTTG
SSIV 45/10 NA     AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGGATTGGTCTTG
SSIV 18/10 NA     AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGGATTGGTCTTG
SSIV 206/09 NA    AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGGATTGGTCTTG
SSIV 270/09 NA    AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGGATTGGTCTTG
SSIV 294/09 NA    AGCAGCATATCCTTTTGTGGTGTAAATAGTGACACTGTGAATTGGTCTTG
Haseluenne 2003   AGCAGCATATCCTTTTGTGGTGTGAATAGTGACACTGTGGGTTGGTCTTG
Mexico 2009       AGCAGCATATCCTTTTGTGGTGTAAACAGTGACACTGTGGGTTGGTCTTG
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SSIV 54/10 NA    GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
SSIV 132/09 NA   GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
SSIV 199/09 NA   GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
SSIV 247/09 NA   GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
SSIV 45/10 NA    GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
SSIV 18/10 NA    GCCAGACGGTGCTGAGTTGCCATTTACAATTGACAAGTAATTTGTTCAAA
SSIV 206/09 NA   GCCAGACGGTGCTGAGTTGCCATTTACAATTGACAAGTAATTTGTTCAAA
SSIV 270/09 NA   GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
SSIV 294/09 NA   GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
Haseluenne 2003  GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCAAA
Mexico 2009      GCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAATTTGTTCA--
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SSIV 54/10 NA    AAACTCCTTGTTTCTACTGATCACGCGT
SSIV 132/09 NA   AAACTCCTTGTTTCTACTGATCACGCGT
SSIV 199/09 NA   AAACTCCTTGTTTCTACTGATCACGCGT
SSIV 247/09 NA   AAACTCCTTGTTTNTACTGATCACGCGT
SSIV 45/10 NA    AAACTCCTTGTTTCTACTGATCACGCGT
SSIV 18/10 NA    AAACTCCTTGTTTCTACNGATCACGCGT
SSIV 206/09 NA   AAACTCCTTGTTTCTACTGATCACGCGT
SSIV 270/09 NA   AAACTCCTTGTTTCTACTKATCACGCGT
SSIV 294/09 NA   AAACTCCTTGTTTCTACNGATCACGCGT
Haseluenne 2003  AAA--------------------------
Mexico 2009      --------------------------
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Legend:
Nucleotide sequences of segment 6. Highlighted in red: mutation (R194G), which increases viral fitness in oseltamivir resistant strains. Highlighted in blue: mutation (H274Y), causing resistance against oseltamivir.

Appendix 3b

Neuraminidase

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SSIV 132/09 NA   MNPNQKIIIISSICLINGITSLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 45/10 NA    MNPNQKIIIISSICLINGITSLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 199/09 NA   MNPNQKIIIISSICLINGITSLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 18/10 NA    MNPNQKIIIISSICLINGIASLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 206/09 NA   MNPNQKIIIISSICLINGIASLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 294/09 NA   MNPNQKIIIISSICLINGIASLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 270/09 NA   MNPNQKIIIISSICLINGIAGLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 54/10 NA    MNPNQKIITISSICLINGITSLILQIGNIISIWISHSIQIGNQNQTETCS
SSIV 247/09 NA   MNPNQXIIIIXSICLINGITSLILQIGNIISIWISHSIQIGNQNQTETCS
Haseluenne 2003 MNPNQKIIIISSICMINGIASLILQIGNIISIWISHSIQIGNQNQTETCN
Mexico 2009      MNPNQKIITIGSVCMTIGMANLILQIGNIISIWISHSIQLGNQNQIETCN
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SSIV 132/09 NA   QNIITYENNTWVNQTYVNISNNNFVAEQAVVSMKLAGSSSLCPVSGWAIY
SSIV 45/10 NA    QNIITYENNTWVNQTYVNISNNNFVAEQAVVSMKLAGSSSLCPVSGWAIY
SSIV 45/10 NA    QNIITYENNTWVNQTYVNISNNNFVAEQAVVSMKLAGSSPLCPVSGWAIY
SSIV 18/10 NA    QNIITYENNTWVNQTYVNISNNNFVAEQAVVSIKLAGSSSLCPVSGWAIY
SSIV 206/09 NA   QNIITYENNTWVNQTYVNISNNNFVAEQAVVSIKLAGSSSLCPVSGWAIY
SSIV 294/09 NA   QNIITYENYTWVNQTYVNISNNNFVAEQAVVSIKLAGSSSLCPVSGWAIY
SSIV 270/09 NA   QNIITYENNTWVNHTYVNISNNNFVAEQAVVSTKLAGSSSLCPVSGWAIY
SSIV 54/10 NA    QNIITYENNTWVNQTYVNISNNNFVAEQAVVSXKLAGSSSLCPVSGWAIY
SSIV 247/09 NA   QNIITYENNTWVNQTYVNISNNNFVAXQAVVSMKLAGXSPLCPVSGWAIY
Haseluenne 2003 QSVITYENNTWVNQTYVNISNNNFVAEQAVVSVKLAGSSPLCSVSGWAIY
Mexico 2009      QSVITYENNTWVNQTYVNISNTNFAAGQSVVSVKLAGNSSLCPVSGWAIY
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SSIV 132/09 NA   SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 45/10 NA    SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 45/10 NA    SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 18/10 NA    SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 206/09 NA   SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 294/09 NA   SKDNSVRVGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 270/09 NA   SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 54/10 NA    SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
SSIV 247/09 NA   SKDNSVRIGSKGDVFVIREPFISCSHLECRTFFLTXGALLNDKHSNGTIK
Haseluenne 2003 SKDNSVRIGSRGDVFVIREPFISCSHLECRTFFLTQGALLNDKHSNGTIK
Mexico 2009      SKDNSVRIGSKGDVFVIREPFISCSPLECRTFFLTQGALLNDKHSNGTIK
                 *******:**:************** ********* **************

SSIV 132/09 NA   DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
SSIV 45/10 NA    DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
SSIV 45/10 NA    DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHNGTSWLTIGISGPDN
SSIV 18/10 NA    DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
SSIV 206/09 NA   DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
SSIV 294/09 NA   DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
SSIV 270/09 NA   DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
SSIV 54/10 NA    DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
SSIV 247/09 NA   DRSPYRTLXSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
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Haseluenne 2003 DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGTSWLTIGISGPDN
Mexico 2009     DRSPYRTLMSCPIGEVPSPYNSRFESVAWSASACHDGINWLTIGISGPDN
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SSIV 132/09 NA  GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPNNGQ
SSIV 45/10 NA   GAVAVLKYNDIITDTIRSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
SSIV 45/10 NA   GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
SSIV 18/10 NA   GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCYTVMTDGPSNGQ
SSIV 206/09 NA  GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
SSIV 294/09 NA  GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
SSIV 270/09 NA  GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
SSIV 54/10 NA   GAVAVLKYNDIITDTIKSWRNNILRTQESECVCVNGSCFTVMTDGPSNGQ
SSIV 247/09 NA  GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
Haseluenne 2003 GAVAVLKYNDIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
Mexico 2009     GAVAVLKYNGIITDTIKSWRNNILRTQESECACVNGSCFTVMTDGPSNGQ
                *********.******:**************.******:*******.***

SSIV 132/09 NA  ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 45/10 NA   ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 45/10 NA   ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 18/10 NA   ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 206/09 NA  ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 294/09 NA  ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 270/09 NA  ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 54/10 NA   ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
SSIV 247/09 NA  ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDXGEIICVCRDNWXGXN
Haseluenne 2003 ASYKIFKIEKGKVVKSVELNAPNYHYEECSCYPDSGEIICVCRDNWHGSN
Mexico 2009     ASYKIFRIEKGKIVKSVEMNAPNYHYEECSCYPDSSEITCVCRDNWHGSN
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SSIV 132/09 NA  RPWVSFNQNLEYQIGYICSGILGDNPRPNDRTGSCGPVSLNGANGVKGFS
SSIV 45/10 NA   RPWVSFNQNLEYQIGYICSGILGDNPRPNDRTGSCDPVSLNGANGVKGFS
SSIV 45/10 NA   RPWVSFNQNLEYQIGYICSGILGDNPRPNDRTGSCGPVSLNGANGVKGFS
SSIV 18/10 NA   RPWVSFNQNLEYQIGYICSGVLGDNPRPNDRTGSCGPVSLNGANGVKGFS
SSIV 206/09 NA  RPWVSFNQNLEYQIGYICSGVLGDNPRPNDRTGSCGPVSLNGANGVKGFS
SSIV 294/09 NA  RPWVSFNQNLEYQIGYICSGVLGDNPRPNDRTGSCGPVSLNGANGVKGFS
SSIV 270/09 NA  RPWVSFNQNLEYQIGYICSGVLGDNPRPNDRTGSCGPVSLNGANGVKGFS
SSIV 54/10 NA   RPWVSFNQDLEYQIGYICSGILGDNPRPNDRTGSCGPVSLNGANGVKGFS
SSIV 247/09 NA  RPWVSFNQNXEYQIGXICSGILGDNPRPNDRTGSCGPVSLNGANGVKGFS
Haseluenne 2003 RPWVSFNQNLEYQIGYICSGVLGDNPRPNDRTGSCGPVSSNGANGVKGFS
Mexico 2009     RPWVSFNQNLEYQIGYICSGIFGDNPRPNDKTGSCGPVSSNGANGVKGFS
                ********: ***** ****::********:****.*** **********

SSIV 132/09 NA  FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 45/10 NA   FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 45/10 NA   FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 18/10 NA   FKYGNGVWIGRTKSTSSRSGFEMVWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 206/09 NA  FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 294/09 NA  FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 270/09 NA  FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 54/10 NA   FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFLVKQDIVGMTDWS
SSIV 247/09 NA  FKYGNGVWIGRXKSXSSRSGFEMIWDPNGWTGXDNNFLVKQDIVGMTDWS
Haseluenne 2003 FKYGNGVWIGRTKSTSSRSGFEMIWDPNGWTGTDNNFSVKQDIVGITDWS
Mexico 2009     FKYGNGVWIGRTKSISSRNGFEMIWDPNGWTGTDNNFSIKQDIVGINEWS
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SSIV 132/09 NA  GYSGSFVQHPELTGMNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 45/10 NA   GYSGSFVQHPELTGMNCIRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 45/10 NA   GYSGSFVQHPELTGMNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 18/10 NA   GYSGSFVQHPELTGMNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 206/09 NA  GYSGSFVQHPELTGMNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 294/09 NA  GYSGSFVQHPELTGMNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 270/09 NA  GYSGSFVQHPELTGMNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 54/10 NA   GYSGSFVQHPELTGMNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
SSIV 247/09 NA  GYSGSFVQHPELTGMNXMRPXXWVELIRGRPKEXXIWTSGSSISFXGVNS
Haseluenne 2003 GYSGSFVQHPELTGLNCMRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
Mexico 2009     GYSGSFVQHPELTGLDCIRPCFWVELIRGRPKENTIWTSGSSISFCGVNS
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SSIV 132/09 NA  DTVDWSWPDGAELPFTIDK
SSIV 45/10 NA   DTVDWSWPDGAELPFTIDK
SSIV 45/10 NA   DTVDWSWPDGAELPFTIDK
SSIV 18/10 NA   DTVDWSWPDGAELPFTIDK
SSIV 206/09 NA  DTVDWSWPDGAELPFTIDK
SSIV 294/09 NA  DTVNWSWPDGAELPFTIDK
SSIV 270/09 NA  DTVDWSWPDGAELPFTIDK
SSIV 54/10 NA   DTVNWSWPDGAELPFTIDK
SSIV 247/09 NA  DTVGWSWPDGAELPFTIDK
Haseluenne 2003 DTVGWSWPDGAELPFTIDK
Mexico 2009     DTVGWSWPDGAELPFTIDK
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Legend:

Amino acid sequences of Neuraminidase. R194G highlighted in red; H274Y highlighted in blue.

Appendix 4a

Segment 5

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SSIV 206/09 NP      ACGCGTGATCAGTAGAAACAAGGGTATTTTTTCTTTAACTGTCATACTCC
SSIV 18/10 NP       ACGCGTGATCAGTAGAAACAAGGGTATTTTT-CTTTAACTGTCATACTCC
SSIV 294/09 NP      ACGCGTGATN-GTAGAAACAAGGGTATTTTTTCTTTAACTGTCATACTCC
SSIV 199/09 NP      ACGCGTGATCAGTAGAAACAAGGGTATTTTTTCTTTAACTGTCATACTCC
SSIV 246/09 NP      ACGCGTGAN-AGNANAAACAAGGGTATTTTTTCTTTAATTGTCATACTCC
SSIV 45/10 NP       ACGCGTGATCAGTAGAAACAAGGGTATTTTTTCTTTAACTGTCATAATCC
SSIV 132/09 NP      TCGCGTGATCNGTAGAAACAAGGGTATTTTTTCTTTAACTGTCATACTCT
Haseluenne 2003     ----------AGTAGAAACAAGGGTATTTTT-CTTTAGTTGTCATACTCC
Mexico 2009         ----------------------------------CTCAACTGTCATACTCC
                                                      * *  ******* **

SSIV 206/09 NP      TCTGCATTGTCTCCGAAGAAATAAGATCCTTCATTACTCATGTCAAAGGA
SSIV 18/10 NP       TCTGCATTGTCTCCGAAGAAATAAGATCCTTCATTACTCATGTCAAAGGA
SSIV 294/09 NP      TCTGCATTGTCTCCGAAGAAATAAGATCCTTCATTATTCATGTCAAAGGA
SSIV 199/09 NP      TCTGCATTGTCTCCGAAGAAATAAGATCCTTCATTACTCATGTCAAAGGA
SSIV 246/09 NP      TCTGCATTGTCTCCGAANAAATAAGATCCTTCATTACTCATGTCAAAGGA
SSIV 45/10 NP       TCTGCATTGTCTCCGAAGAAATAAGATCCTTCATTACTCATGTCAAAGGA
SSIV 132/09 NP      TCTGCATTGTCTCCGAAGAAATAAGATCCTTCATTACTCATGTCAAAGGA
Haseluenne 2003     TCTGCATTGTCTCCGAAGAAATAAGATCCTTCATTACTCATGTCAAAGGA
Mexico 2009         TCTGCATTGTCTCCGAAGAAATAAGACCCTTCATTACTCATGTCAAAGGA
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SSIV 206/09 NP      AGGCACGATCGGGTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
SSIV 18/10 NP       AGGCACGATCGGGTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
SSIV 294/09 NP      AGGCACGATCGGGTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
SSIV 199/09 NP      AGGCACGATCGGGTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
SSIV 246/09 NP      AGGCACGATCGGNTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
SSIV 45/10 NP       AGGCACGATCGGGCTCGTTGCCTTTTCGTCCGATAGCTCGAAGACTCCCC
SSIV 132/09 NP      AGGCACGATCGGGTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
Haseluenne 2003     AGGCACGATCGGGTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
Mexico 2009         AGGCACGATCGGGTTCGTTGCCTTTTCGTCCGAGAGCTCGAAGACTCCCC
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SSIV 206/09 NP      GCCCCTGGAAGGACACATCTTCTGGTCTGGCACTTTCCATCATTCTTATA
SSIV 18/10 NP       GCCCCTGGAAGGACACATCTTCTGGTCTGGCACTTTCCATCATTCTTATA
SSIV 294/09 NP      GCCCCTGGAAGGACACATCTTCTGGTCTGGCACTTTCCATCATTCTTATA
SSIV 199/09 NP      GCCCCTGGAAGGACACATCTTCTGGTCTGGCACTTTCCATCATTCTTATA
SSIV 246/09 NP      GCCCCTGGAAGGACACATNTTCTGGTCTGGCACTTTCCATCATTCTTATA
SSIV 45/10 NP       GCCCCTGGAAGGACACATCTTCTGGTCTGGCACTTTCCATCATTTTTATA
SSIV 132/09 NP      GCCCCTGGAAGGACACATCTTCTGGTCTGGCACTTTCCATCATTCTTATA
Haseluenne 2003     GCCCTTGGAAGGACACATCTTCTGGTCTGGCACTTTCCATCATTCTTATA
Mexico 2009         GCCCCTGGAAGGACAAATCTTCTGGCTTTGCACTTTCCATCATTCTTATA
                    **** ********** ** ******  * *************** *****

SSIV 206/09 NP      ATTTCAGTCCTCATATCAGATGTTCTGCCTTCAGTGTTTCCAGTGAATGC
SSIV 18/10 NP       ATTTCAGTCCTCATATCAGATGTTCTGCCTTCAGTGTTTCCAGTGAATGC
SSIV 294/09 NP      ATTTCAGTCCTCATATCAGATGTTCTGCCTTCAGTGTTTCCAGTGAATGC
SSIV 199/09 NP      ATTTCAGTCCTCATATCAGAAGTTCTGCCTTCAGTGTTTCCAGTGAACGC
SSIV 246/09 NP      ATTTCAGTCCTCATATCAGAAGTTCTGCCTTCAGTGTTTCCAGNGAACGC
SSIV 45/10 NP       ATTTCAGTCCTCATATCAGATGTTCTGCCTTCAGTGTTTCCAGTGAACGC
SSIV 132/09 NP      ATTTCAGTCCTCATATCAGATGTTCTGCCTTCAGTGTTTCCAGTGAACGC
Haseluenne 2003     ATTTCAGTCCTCATATCAGATGTTCTGCCTTCAGTGTTTCCAGTAAATGC
Mexico 2009         ACTTCTGTTCGCATGTCGGATGTCCGTCCTTCATTGTTCCCGCTGAATGC
                    * *** ** * *** ** ** ** * ******* **** **    ** **

SSIV 206/09 NP      TGCCATGATGGTCGCTCTCTCAAAAGGAAGATTTCTCTGTACTGAGAAAG
SSIV 18/10 NP       TGCCATGATGGTCGCTCTCTCAAAAGGAAGATTTCTCTGTACTGAGAAAG
SSIV 294/09 NP      TGCCATGATGGTCGCTCTCTCAAAAGGAAGATTTCTCTGTACTGAGAAAG
SSIV 199/09 NP      TGCCATGATGGTCGCTCTCTCAAAAGGAAGATTTCTCTGTACTGAGAAAG
SSIV 246/09 NP      TGCCATGATGGTCGCTCTCTCAAAAGGAAGATTTNNCTGTNCTGAGAAAG
SSIV 45/10 NP       TGCCATGATAGTCGCTCTCTCAAAAGGAAGATTTCTTTGTACTGAGAAAG
SSIV 132/09 NP      TGCCATGATGGTTGCTCTCTCAAAAGGAAGATTTCTCTGTACTGAGAAAG
Haseluenne 2003     TGCCATGATGGTAGCTCTCTCGAAAGGAAGATTTCTCTGTACTGAGAAAG
Mexico 2009         TGCCATAACGGTTGCTCTTTCAAAAGGGAGATTCCGCTGCACTGAGAATG
                    ****** *  ** ***** ** ***** *****    **  ******* *

SSIV 206/09 NP      TAGGTTGTACACTGATTTGCCCTGCGGATGCTCTCTGTTGATTGGTGTTC
SSIV 18/10 NP       TAGGTTGTACACTGATTTGCCCTGCGGATGCTCTCTGTTGATTGGTGTTC
SSIV 294/09 NP      TAGGTTGTACACTGATTTGCCCTGCGGATGCTCTCTGTTGATTGGTGTTC
SSIV 199/09 NP      TAGGTTGTACACTGATTTGCCCAGCGGATGCTCTCTGTTGATTGGTGTTC
SSIV 246/09 NP      TAGGTTGTSCACTGATTTGCCCTGCGGATGCTCTCTGTTGATTGGTGTTC
SSIV 45/10 NP       TAGGTTGTACACTGATTTGCCCTGCGGATGCTCTCTGTTGATTGGTGTTC
SSIV 132/09 NP      TAGGTTGTACACTGATTTGCCCTGCGGATGCTCTCTGTTGATTGGTATTC
Haseluenne 2003     TAGGTTGTACACTGATTTGCCCTGCGGATGCTCTCTGTTGATTGGTGTTT
Mexico 2009         TAGGCTGCACACTGATCTGGCCTGCGGATGCCTTTTGTTGATTGGTATTT
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SSIV 206/09 NP      CCTCCGCTCCTGGTTCTTATAGCCCAGTATTTGCTTCTAAGTTCAAGAGT
SSIV 18/10 NP       CCTCCGCTCCTGGTTCTTATAGCCCAGTATTTGCTTCTAAGTTCAAGAGT
SSIV 294/09 NP      CCTCCGCTCCTGGTTCTTATAGCCCAGTATTTGCTTCTAAGTTCAAGAGT
SSIV 199/09 NP      CCTCCGCTCCTGGTTCTTATAGCCCAGTATTTGCTTCTAAGTTCAAGAGT
SSIV 246/09 NP      CCTCCGNTCCTGGTTCTTATAGCCCAGTNTTTGCTTCTGAGTNCAAGAGT
SSIV 45/10 NP       CCTCCGCTCCTGGTTCTTATAGCCCAGTATTTGCTTCTAAGTTCAAGAGT
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SSIV 132/09 NP      CCTCCGCTCCTGGTTCTTATAGCCCAGTATTTGCTTCTAAGTTCAAGAGT
Haseluenne 2003     CCTCCGCTCCTGGTTCTTATAGCCCAGTATTTGCTTCTAAGTTCAAGAGT
Mexico 2009         CCTCCACTCCTGGTCCTTATGGCCCAGTATCTGCTTCTCAGTTCCAGGGT
                    *****  ******* ***** ******* * ******* *** * ** **

SSIV 206/09 NP      AATGGAGTCCATTGTTTCCATGTTTTCATTTGAAGCAATTTGAATCCCTC
SSIV 18/10 NP       AATGGAGTCCATTGTTTCCATGTTTTCATTTGAAGCAATTTGAATCCCTC
SSIV 294/09 NP      AATGGAGTCCATTGTTTCCATGTTTTCATTTGAAGCAATTTGAACCCCTC
SSIV 199/09 NP      AATGGAGTCCATTGTTTCCATGTTTTCATTTGAAGCAATTTGAACCCCTC
SSIV 246/09 NP      AATGGAGTCCATTGTTTCCATGTTTTCATTTGAAGCAATTTGAACCCCTC
SSIV 45/10 NP       AATGGAGTCCATTGTTTCCATGTTTTCATTTGAAGCAATTTGAACCCCTC
SSIV 132/09 NP      AATGGAGTCCATTGTTTCCATGTTTTCATTTGAAGCAATTTGAACCCCTC
Haseluenne 2003     AATGGAGTCCATTGTTTCCATGTTTTCATTCGAAGCAATTTGAACCCCTC
Mexico 2009         ATTGGAGTCCATGATTTCCACATTCTCATTTGAAGCAATCTGGACCCCTC
                    * **********  ******  ** ***** ******** ** * *****

SSIV 206/09 NP      TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCTGATAAAACTT
SSIV 18/10 NP       TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCTGATAAAACTT
SSIV 294/09 NP      TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCTGATAAAACTT
SSIV 199/09 NP      TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCTGATAAAACTT
SSIV 246/09 NP      TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCNGATAAAACTT
SSIV 45/10 NP       TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCTGATAAAACTT
SSIV 132/09 NP      TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCTGATAAAACTT
Haseluenne 2003     TGGTGGACAATTGTCCTCTTGGGATCACTTTTGTCCCTCTGATAAAACTT
Mexico 2009         TTGTGGAAAGCTTTCCTCTTGGAATCACTTTCTTTCCTCTTATGAAACTT
                    * ***** *  * ********* ********  * ****  ** ******

SSIV 206/09 NP      GACACCCTCAGATCTTCAAATGCTGCAGAATGGCATGCCATCCATACCAG
SSIV 18/10 NP       GACACCCTCAGATCTTCAAATGCTGCAGAATGGCATGCCATCCATACCAG
SSIV 294/09 NP      GACACCCTCAGATCTTCAAATGCTGCAGAATGGCATGCCATCCATACCAG
SSIV 199/09 NP      GACACCCTCAGATCTTCAAATGCTGCAGAATGGCATGCCATCCATACCAG
SSIV 246/09 NP      GACACCCTCAGATCTTCAAATGCTGCAGAATGGCATGCCATCCATACCAG
SSIV 45/10 NP       GACACCCTCAAATCTTCAAATGCTGCAGAATGGCATGCCATCCATACCAG
SSIV 132/09 NP      GACACCCTCAGATCTTCAAATGCTGCAGAATGGCATGCCATCCATACCAG
Haseluenne 2003     GACACCCTCAGATCTTCAAATGCTGCAGAATGGCATGCCATCCATATCAG
Mexico 2009         GATACTCTTAAATCTTCAAATGCAGCAGAGTGGCATGCCATCCACACCAA
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SSIV 206/09 NP      CTGACTCTTATGGACTGGATTCTCATTTGGTCTAATGAGGCTGAACACTT
SSIV 18/10 NP       CTGACTCTTATGGACTGGATTCTCATTTGGTCTAATGAGGCTGAACACTT
SSIV 294/09 NP      CTGACTCTTATGGACTGGATTCTCATTTGGTCTAATGAGGCTGAACACTT
SSIV 199/09 NP      CTGACTCTTATGGACTGGATTCTCATTCGGTCTAATGAGGCTGAACACTT
SSIV 246/09 NP      CTGACTCTTATGGACCGNATTCTCATTCGGTCTAATGAGGCTGAACACTT
SSIV 45/10 NP       CTGACTCTTATGGACTGGATTCTCATTCGGTCTAATGAGGCTGAATACTT
SSIV 132/09 NP      CTGACTCTTATGGACTGGATTCTCATTTGGTCTAATGAGGCTGAACACTT
Haseluenne 2003     CTGACTCTTATGGACTGGATTCTCATTTGGTCTAATGAGGCTGAACACCT
Mexico 2009         TTGACTCTTGTGAGCTGGGTTTTCATTTGGTCTCATCAGGCTGACCACTT
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SSIV 206/09 NP      GGCTGTTCTGAAGCAGACGAAAAGGATCTATCCCGACTAGGGAGTACCCT
SSIV 18/10 NP       GGCTGTTCTGAAGCAGACGAAAAGGATCTATCCCGACTAGGGAGTACCCT
SSIV 294/09 NP      GGCTGTTCTGAAGCAGACGAAAAGGATCTATCCCGACTAGGGAGTACCCT
SSIV 199/09 NP      GGCTGTTCTGAAGCAGACGAAAAGGATCTACCCCGACTAGGGAGTACCCT
SSIV 246/09 NP      GGCTGTTCTGAAGCAGACNAAAAGGATCTACCCCGACTAGGGAGTACCCT
SSIV 45/10 NP       GGCTGTTCTGAAGCAGACGAAAAGGATCTACCCCGACTAGGGAGTACCCT
SSIV 132/09 NP      GGCTGTTCTGAAGCAGACGAAAAGGATCTACCCCGACTAGGGAGTACCCT
Haseluenne 2003     GGCTGTTCTGAAGCAGACGAAAAGGATCTATTCCGACTAGGGAGTACCCT
Mexico 2009         GGCTGTTTTGGAGTAATTTGAATGGGTCTATCCCGACCAGTGAGTACCCT
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SSIV 206/09 NP      TCTCTTTCAAAGTCATATCCACTTGCCACAATAAGTCCATATACACAAGC
SSIV 18/10 NP       TCTCTTTCAAAGTCATATCCACTTGCCACAATAAGTCCATATACACAAGC
SSIV 294/09 NP      TCTCTTTCAAAGTCATATCCACTTGCCACAATAAGTCCATATACACAAGC
SSIV 199/09 NP      TCTCTTTCAGAGTCATATCCACTTGCCACAATTAGTCCATATACACAAGC
SSIV 246/09 NP      TCTCTTTCAAAGTCATATCCACTTGNCACAATGAGTCCATATACACAAGC
SSIV 45/10 NP       TCTCTTTCAAAGTCATATCCACTTGCCACAATGAGTCCATATACACAAGC
SSIV 132/09 NP      TCTCTTTCAAAGTCATATCCACTTGCCACAATGAGTCCATATACACAAGC
Haseluenne 2003     TCTCTTTCAAAGTCATATCCACTTGCCACAATAAGTCCATATACGCAAGC
Mexico 2009         TCCCTTTCAAAGTCATGCCCACTTGCTACTGCAAGCCCATACACACAAGC
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SSIV 206/09 NP      AGGCAGGCAGGATTTGTGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
SSIV 18/10 NP       AGGCAGGCAGGATTTGTGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
SSIV 294/09 NP      AGGCAGGCAGGATTTGTGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
SSIV 199/09 NP      AGGCAGGCAGGATTTGTGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
SSIV 246/09 NP      AGGCAGGCAGGATTTGTGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
SSIV 45/10 NP       AGGCAGGCAGGATTTGTGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
SSIV 132/09 NP      AGGCAGACAGGATTTGTGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
Haseluenne 2003     AGGCAGGCAGGATTTATGAGCCACTGATCCTCTCAGAATGAGTGCAGATC
Mexico 2009         AGGCAGGCAGGATTTATGTGCAACTGATCCCCTCAGAATGAGTGCTGACC
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SSIV 206/09 NP      GTGCCAGAAAGATGAGATCTTCAATCTCAGCATTTCCTGGATTTCTGCTT
SSIV 18/10 NP       GTGCCAGAAAGATGAGATCTTCAATCTCAGCATTTCCTGGATTTCTGCTT
SSIV 294/09 NP      GTGCCAGAAAGATGAGATCTTCAATCTCAGCATTTCCTGGATTTCTGCTT
SSIV 199/09 NP      GTGCCAGAAAGATGAGATCTTCAATCTCAGCATTCCCTGGATTTCTGCTT
SSIV 246/09 NP      GTGCCAGAAAGATGAGATCTTCAATCTCAGCATTCCCTGGATTTCTGCTT
SSIV 45/10 NP       GTGCCAGAAAGATGAGATCTTCAATCTCAGCGTTCCCTGGATTTCTGCTT
SSIV 132/09 NP      GTGCCAGAAAGATGAGATCTTCAATCTCAGCATTCCCTGGATTTCTGCTT
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Haseluenne 2003     GTGCCAGAAAAATGAGATCTTCAATCTCAGCATTTCCTGGATTTCTGCTT
Mexico 2009         GTGCCAGGAAAATGAGGTCTTCAATCTCAGCGTTTCCTGGGTTTCGACTT
                    ******* ** ***** ************** ** ***** ****  ***

SSIV 206/09 NP      TCTCGCACCTGGTCCATCATTGCCCGTTGCGCTGCTGTTTGGAATTTCCC
SSIV 18/10 NP       TCTCGCACCTGGTCCATCATTGCCCGTTGCGCTGCTGTTTGGAATTTCCC
SSIV 294/09 NP      TCTCGCACCTGGTCCATCATTGCCCGTTGCGCTGCTGTTTGGAATTTCCC
SSIV 199/09 NP      TCTCGCACCTGGTCCATCATTGCTCGTTGCGCTGCTGTTTGGAATTTCCC
SSIV 246/09 NP      TCTCGCACCTGGTCCGTCATTGCTCNTTGCGCTGCTGTTTGGAATTTTCC
SSIV 45/10 NP       TCTCGCACCTGGTCCATCATTGCTCGTTGCGCTGCTGTTTGGAATTTCCC
SSIV 132/09 NP      TCTCGCACCTGGTCCATCATTGCTCGTTGCGCTGCTGTTTGGAATTTCCC
Haseluenne 2003     TCTCGCACCTGGTCCATCATTGCTCGTTGCGCTGCTGTCTGGAATTTCCC
Mexico 2009         TCTCTTACTTGATCCATCATTGCCCTCTGGGCAGCTGTTTGAAATTTTCC
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SSIV 206/09 NP      TTTGAGGATGTTGCACATTCTCTCATATGCAATTCTTGTTCTCCGTCCAT
SSIV 18/10 NP       TTTGAGGATGTTGCACATTCTCTCATATGCAATTCTTGTTCTCCGTCCAT
SSIV 294/09 NP      TTTGAGGATGTTGCACATTCTCTCATATGCAATTCTTGTTCTCCGTCCAT
SSIV 199/09 NP      TTTGAGGATGTTGCACATTCTCTCATATGCAATTCTTGTTCTTCGTCCAT
SSIV 246/09 NP      TTTGAGGATGTTGCACATTCTCTCATATGCAATTCTTGTTCTTCGTCCAT
SSIV 45/10 NP       TTTGAGGATGTTGCACATTCTCTCATATGCAATTCTTGTTCTTCGTCCAT
SSIV 132/09 NP      TTTGAGGATGTTGCACATTCTTTCATATGCAATTCTTGTTCTTCGTCCAT
Haseluenne 2003     TTTGAGGATGTTGCACATTCTCTCATATGCAATTCTTGTTCTTCGTCCAT
Mexico 2009         TTTGAGGATATTGCACATTCTTTCATAAGCAACCCTTGTCCTTCGTCCAT
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SSIV 206/09 NP      TCTCGCCTCTCCAGAAGTTCCGATCATTGATACCCCGCTTTATCATTCGA
SSIV 18/10 NP       TCTCGCCTCTCCAGAAGTTCCGATCATTGATACCCCGCTTTATCATTCGA
SSIV 294/09 NP      TCTCGCCTCTCCAGAAGTTCCGATCATTGATACCCCGCTTTATCATTCGA
SSIV 199/09 NP      TCTCGCCTCTCCAGAAGTTTCTATCATTGATACCCCGCTTTATCATTCGA
SSIV 246/09 NP      TCTCGCCTCTCCAGAAGTTTCGATCATTGATACCCCGCTTTATCATTCGA
SSIV 45/10 NP       TCTCGCCTCTCCAGAAGTTTCGATCATTGATACCCCGCTTTATCATTCGA
SSIV 132/09 NP      TCTCGCCTCTCCAGAAGTTTCGATCATTGATACCCCGCTTTATCATTCGA
Haseluenne 2003     TCTCGCCTCTCCAGAAGTTCCGATCATTGATACCCCGCTTTATCATTCGA
Mexico 2009         TTTCACCCCTCCAGAAATTTCGGTCATTGATTCCACGTTTGATCATTCTG
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SSIV 206/09 NP      ATCAGCTCCATTACTAGTGTCCCAACTCCCTTTACTGCTGCACCAGCAGC
SSIV 18/10 NP       ATCAGCTCCATTACTAGTGTCCCAACTCCCTTTACTGCTGCACCAGCAGC
SSIV 294/09 NP      ATCAGCTCCATTACTAGTGTCCCAACTCCCTTTACTGCTGCACCAGCAGC
SSIV 199/09 NP      ATCAGCTCCATTACTAGTGTCCCAACTCCCTTTACTGCTGCACCAGCAGC
SSIV 246/09 NP      ATCAGCTCCATTACTAGTGTCCCAACTCCCTTTACTGCTGCACCAGCAGC
SSIV 45/10 NP       ATCAGCTCCATTACTAGTGTCCCAACTCCCTTTACTGCTGCACCAGCAGC
SSIV 132/09 NP      ATCAGCTCCATTACTAGTGTCCCAACTCCCTTTACTGCTGCACCAGCAGC
Haseluenne 2003     ATCAGCTCCATTACCACTGTTCCAACTCCCTTTACTGCTGCACCAGCAGC
Mexico 2009         ATTAACTCCATTGCTATTGTTCCAACTCCTTTCACCGCAGCACCTGCGGC
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SSIV 206/09 NP      TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGAGAGCACATTCTGG
SSIV 18/10 NP       TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGAGAGCACATTCTGG
SSIV 294/09 NP      TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGAGAGCACATTCTGG
SSIV 199/09 NP      TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGAGAGCACATTCTGG
SSIV 246/09 NP      TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGAGAGCACATTCTGG
SSIV 45/10 NP       TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGAGAGCACATTCTGG
SSIV 132/09 NP      TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGGGAGCACATTCTGG
Haseluenne 2003     TCCAGATCTCCTTGGGAGAGTTGAACCTTGCATCAGAGAGCACATTCTGG
Mexico 2009         ACCAGACCTTCTGGGAAGTGTTGAACCTTGCATTAGAGAGCACATTCTGG
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SSIV 206/09 NP      GATCCATTCCAGTACGCACTAAAGCTCTTGTTCTCTGATATGTGGCATCA
SSIV 18/10 NP       GATCCATTCCAGTACGCACTAAAGCTCTTGTTCTCTGATATGTGGCATCA
SSIV 294/09 NP      GATCCATTCCAGTACGCACTAAAGCTCTTGTTCTCTGATATGTGGCATCA
SSIV 199/09 NP      GATCCATTCCAGTACGCACTAAAGCTCTTGTTCTCTGATATGTGGCATCA
SSIV 246/09 NP      GATCCATTCCAGTACGCACTAAAGCTCTTGTTCTCTGATATGTGGCATCA
SSIV 45/10 NP       GATCCATTCCAGTACGCACTAAAGCTCTTGTTCTCTGATATGTGGCATCA
SSIV 132/09 NP      GATCCATTCCAGTACGCACTAAAGCTCTTGTTCTCTGATATGTGGCATCA
Haseluenne 2003     GATCCATCCCAGTACGCACTAAAGCTCTTGTTCTCTGATACGTGGCATCA
Mexico 2009         GATCCATTCCGGTGCGAACAAGCGCTCTTGTTCTCTGATATGTGGCATCA
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SSIV 206/09 NP      TTCAGGTTGGAATGCCAAATCATCAGGTGAGTAAGACCAGCAGTAGCATC
SSIV 18/10 NP       TTCAGGTTGGAATGCCAAATCATCAGGTGAGTAAGACCAGCAGTAGCATC
SSIV 294/09 NP      TTCAGGTTGGAATGCCAAATCATCAGGTGAGTAAGACCAGCAGTAGCATC
SSIV 199/09 NP      TTCAGGTTGGAATGCCAAATCATCAGGTGAGTGAGACCAGCAGTAGCATC
SSIV 246/09 NP      TTCAGGTTGGAATGCCAAATCATCAGGTGAGTGAGACCAGCAGTAGCATC
SSIV 45/10 NP       TTCAGGTTGGAATGCCAAATCATCAGGTGAGTGAGACCAGCAGTAGCATC
SSIV 132/09 NP      TTCAGGTTGGAATGCCAAATCATCAGGTGAGTGAGACCAGCAGTAGCATC
Haseluenne 2003     TTCAGGTTGGAATGCCAAATCATCAGGTGAGTGAGACCAGCAGTAGCATC
Mexico 2009         TTCAGGTTGGAATGCCAAATCATGATATGAGTAAGACCTGCTGTTGCATC
                    ********************** *  ***** ***** ** ** *****

SSIV 206/09 NP      TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTGATTTCATCTTTGTCAT
SSIV 18/10 NP       TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTGATTTCATCTTTGTCAT
SSIV 294/09 NP      TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTGATTTCATCTTTGTCAT
SSIV 199/09 NP      TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTGATTTCATCTTTGTCAT
SSIV 246/09 NP      TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTGATTTCATCTTTGTCAT
SSIV 45/10 NP       TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTGATTTCATCTTTGTCAT
SSIV 132/09 NP      TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTAATTTCATCTTTGTCAT
Haseluenne 2003     TTCACCATTGTTTGCTTGGCGCCAAATTCTCCTGATTTCATCTTTGTCAT
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Mexico 2009         TTCGCCATTGTTTGCTTGGCGCCAAACTCTCCTTATTTCTTCTTTGTCAT
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SSIV 206/09 NP      ACAGAATCAGTTCTCTCATCCATTTTCCATCTCTCTTTTTGTAGATTGGA
SSIV 18/10 NP       ACAGAATCAGTTCTCTCATCCATTTTCCATCTCTCTTTTTGTAGATTGGA
SSIV 294/09 NP      ACAGAATCAGTTCTCTCATCCATTTTCCATCTCTCTTTTTGTAGATTGGA
SSIV 199/09 NP      ACAGAATCAGTTCTCTCATCCATTTTCCATCTCTCTTTTTGTAGATTGGA
SSIV 246/09 NP      ACAGAATCAGTTCTCTCATCCATTTTCCATCTCTCTTTTTGTAGATTGGA
SSIV 45/10 NP       ACAGAATCAGTTCTCTCATCCATTTTCCATCTCTCTTTTTGTAGATTGGA
SSIV 132/09 NP      ACAGAATCAGTTCTCTCATCCATTTTCCATCTCTCTTTTTGTAGATTGGA
Haseluenne 2003     ACAGAATCAGCTCTCTCATCCATTTTCCGTCTCTCCTTTTGTAGATTGGA
Mexico 2009         AAAGGATGAGTTCTCTCATCCACTTTCCGTCTACTCTTCTATATATGGGT
                    * ** ** ** ********** ***** ***    ** * ** ** **

SSIV 206/09 NP      CCTCCAGTTTTCTTTGGATCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
SSIV 18/10 NP       CCTCCAGTTTTCTTTGGATCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
SSIV 294/09 NP      CCTCCAGTTTTCTTTGGATCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
SSIV 199/09 NP      CCTCCAGTTTTCTTTGGATCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
SSIV 246/09 NP      CCTCCAGTTTTCTTTGGGTCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
SSIV 45/10 NP       CCTCCAGTTTTCTTTGGATCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
SSIV 132/09 NP      CCTCCAGTTTTCTTTGGATCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
Haseluenne 2003     CCTCCAGTTTTCTTTGGATCTTTCCCCGCACTGGGATGTTCTTCCAAGTA
Mexico 2009         CCTCCTGTTTTCTTAGGGTCCTTCCCAGCACTGGGATGCTCTTCTAGGTA
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SSIV 206/09 NP      TTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
SSIV 18/10 NP       CTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
SSIV 294/09 NP      TTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
SSIV 199/09 NP      TTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
SSIV 246/09 NP      TTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
SSIV 45/10 NP       TTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
SSIV 132/09 NP      TTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
Haseluenne 2003     TTTGTTTCTCCTCTCGTCAAATGCAGAGAGGACCATTCTCTCTATCGTTA
Mexico 2009         TTTATTTCTTCTCTCATCAAAAGCAGAAAGCACCATCCTCTCTATTGTTA
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SSIV 206/09 NP      TACTATTTTGGATCAGCCTCCCTTCATAGTCACTGAGTTGGAGTTCAGTA
SSIV 18/10 NP       TACTATTTTGGATCAGCCTCCCTTCATAGTCACTGAGTTGGAGTTCAGTA
SSIV 294/09 NP      TACTATTTTGGATCAGCCTCCCTTCATAGTCACTGAGTTGGAGTTCAGTA
SSIV 199/09 NP      TACTATTTTGGATCAGTCTCCCTTCATAGTCACTGAGTTGGAGTTCAGTA
SSIV 246/09 NP      TGCTATTTTGGATCAGTCTCCCTTCATAGTCACTGAGTTGGAGTTCAGTA
SSIV 45/10 NP       TACTATTTTGGATCAGTCTCCCTTCATAGTCACTGAGTTGGAGTTCAGTA
SSIV 132/09 NP      TACTATTTTGGATCAGTCTCCCTTCATAGTCACTGAGTTGGAGTTCAGTA
Haseluenne 2003     TACTATTTTGGATCAGCCTCCCTTCATAGTCACCGAGTTGGAGTTCAGTA
Mexico 2009         TGCTATTCTGGATTAGTCGTCCATCATAATCACTGAGTTTGAGTTCAGTG
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SSIV 206/09 NP      CACATCTGTATGTAGAATCGTCCAATTCCTTCAACCATTCTCCCAACAGA
SSIV 18/10 NP       CACATCTGTATGTAGAATCGTCCAATTCCTTCAACCATTCTCCCAACAGA
SSIV 294/09 NP      CACATCTGTATGTAGAATCGTCCAATTCCTTCAACCATTCTCCCAACAGA
SSIV 199/09 NP      CACATCTGTATGTAGAATCGTCCAATTCCTTCAACCATTCTCCCAACAGA
SSIV 246/09 NP      CACATCTGTATGTAGAATCGTCCAATTCCTTCAACCATTCTCCCAACAGA
SSIV 45/10 NP       CACATCTGTATGTAGAATCGTCCAATTCCTTCAACCATTCTCCCAACAGA
SSIV 132/09 NP      CACATCTGTATGTAGAATCGTCCAATTCCTTCAACCATTCTCCCAACAGA
Haseluenne 2003     CACATCTGTATGTAGAATCGTCCAATTCCTCCAACCATTCTCCCAACAGA
Mexico 2009         CACATTTGGATGTAGAATCTCCCGATTCCACCAATCATTCTTCCGACAGA
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SSIV 206/09 NP      TGCTCTGATTTCAGTGGCATTTTGGCGTTCTCCACCAGTCTCCATCTGTT
SSIV 18/10 NP       TGCTCTGATTTCAGTGGCATTTTGGCGTTCTCCACCAGTCTCCATCTGTT
SSIV 294/09 NP      TGCTCTGATTTCAGTGGCATTTTGGCGTTCTCCACCAGTCTCCATCTGTT
SSIV 199/09 NP      TGCTCTGATTTCAGTAGCATTTTGGCGTTCTCCACCAGTTTCCATCTGTT
SSIV 246/09 NP      TGCTCTGATTTCAGTAGCATTTTGGCGTTCTCCACCAGTTTCCATCTGTT
SSIV 45/10 NP       TGCTCTGATTTCAGTAGCATTTTGGCGTTCTCCACCAGTTTCCATCTGTT
SSIV 132/09 NP      TGCTCTGATTTCAGTAGCATTTTGGCGTTCTCCACCAGTTTCCATCTGTT
Haseluenne 2003     TGCTCTGATTTCAGTAGCATTTTGGCGTTCTCCACCAGTTTCCATTTGTT
Mexico 2009         TGCTCTGATTTCTGTGGCATCCTGGCGCTCCCCACCAGTCTCCATTTGTT
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SSIV 206/09 NP      CATAAGATCGTTTGGTGCCTTGAGACGCCATGGTGTGAATGTCACTCAGT
SSIV 18/10 NP       CATAAGATCGTTTGGTGCCTTGAGACGCCATGGTGTAAATGTCACTCAGT
SSIV 294/09 NP      CATAAGATCGTTTGGTGCCTTGAGACGCCATGGTGTAAATGTCACTCAGT
SSIV 199/09 NP      CATAAGATCGTTTGGTGCCCTGAGACGCCATGGTGTAAATGTCACTCAGT
SSIV 246/09 NP      CATAAGATCGTTTGGTGCCTTGAGACGCCATGGTGTAAATGTCACTCAGT
SSIV 45/10 NP       CATAAGATCGTTTGGTGCCTTGAGACGCCATGATGTAAATGTCACTCAGT
SSIV 132/09 NP      CATAAGATCGTTTGGTGCCTTGAGACGCCATGGTGTAAATGTCACTCAGT
Haseluenne 2003     CATAAGATCGTTTGGTGCCTTGAGACGCCATGGTGTGAATGTCACTCAGT
Mexico 2009         CATATGATCGTTTGGTGCCTTGAGACGCCATGGCTTCGATGTCACTCATT
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SSIV 206/09 NP      GAGTGATTATCTACCCTGCTTTTGCTGATCACGCGT
SSIV 18/10 NP       GAGTGATTATCTACCCTGCTTTTGCTGATCACGCGT
SSIV 294/09 NP      GAGTGATTATCTACCCTGCTTTTGCTGATCACGCGT
SSIV 199/09 NP      GAGTGATTATCTACCCTGCTTTTGCTGATCACGCGT
SSIV 246/09 NP      GAGTGATTATCTACCCTGCTTTTGCTGATCACGCGT
SSIV 45/10 NP       GAGTGATTATCTACCCTGCTTTTGCTGATCACGCGT
SSIV 132/09 NP      GAGTGATTATCTACCCTGCTTTTGCTGATCACGCGT
Haseluenne 2003     GAGTGATTATCTACCCTGCTTTCGCT----------
Mexico 2009         GAGTGA------------------------------
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Legend:
Nucleotide sequences of segment 5. Highlighted in red: sequence encoding for an immunodominant protective epitope at position 366-374 of the aa sequence. Differences in the pandemic strain Mexico 2009 are highlighted in yellow. Highlighted in blue: 301 V/I and 363 V/I variants in Swiss isolates

Appendix 4b

Nucleoprotein

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SSIV 246/09 NP      MASQGTKRSYEQMETGGERQNATEIRASVGRMVEGIGRFYIQMCTELQLS
SSIV 45/10 NP       MASQGTKRSYEQMETGGERQNATEIRASVGRMVEGIGRFYIQMCTELQLS
SSIV 132/09 NP      MASQGTKRSYEQMETGGERQNATEIRASVGRMVEGIGRFYIQMCTELQLS
SSIV 199/09 NP      MASQGTKRSYEQMETGGERQNATEIRASVGRMVEGIGRFYIQMCTELQLS
SSIV 18/10 NP       MASQGTKRSYEQMETGGERQNATEIRASVGRMVEGIGRFYIQMCTELQLS
SSIV 206/09 NP      MASQGTKRSYEQMETGGERQNATEIRASVGRMVEGIGRFYIQMCTELQLS
SSIV 294/09 NP      MASQGTKRSYEQMETGGERQNATEIRASVGRMVEGIGRFYIQMCTELQLS
Haseluenne 2003     MASQGTKRSYEQMETGGERQNATEIRASVGRMVGGIGRFYIQMCTELQLG
Mexico 2009         MASQGTKRSYEQMETGGERQDATEIRASVGRMIGGIGRFYIQMCTELKLS
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SSIV 246/09 NP      DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKKR
SSIV 45/10 NP       DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKKR
SSIV 132/09 NP      DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKKR
SSIV 199/09 NP      DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKKR
SSIV 18/10 NP       DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKKR
SSIV 206/09 NP      DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKKR
SSIV 294/09 NP      DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKKR
Haseluenne 2003     DYEGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYKRR
Mexico 2009         DYDGRLIQNSITIERMVLSAFDERRNKYLEEHPSAGKDPKKTGGPIYRRV
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SSIV 246/09 NP      DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
SSIV 45/10 NP       DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
SSIV 132/09 NP      DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
SSIV 199/09 NP      DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
SSIV 18/10 NP       DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
SSIV 206/09 NP      DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
SSIV 294/09 NP      DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
Haseluenne 2003     DGKWMRELILYDKDEIRRIWRQANNGEDATAGLTHLMIWHSNLNDATYQR
Mexico 2009         DGKWMRELILYDKEEIRRVWRQANNGEDATAGLTHIMIWHSNLNDATYQR
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SSIV 246/09 NP      TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTLVMELIRMIKRG
SSIV 45/10 NP       TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTLVMELIRMIKRG
SSIV 132/09 NP      TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTLVMELIRMIKRG
SSIV 199/09 NP      TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTLVMELIRMIKRG
SSIV 18/10 NP       TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTLVMELIRMIKRG
SSIV 206/09 NP      TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTLVMELIRMIKRG
SSIV 294/09 NP      TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTLVMELIRMIKRG
Haseluenne 2003     TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTVVMELIRMIKRG
Mexico 2009         TRALVRTGMDPRMCSLMQGSTLPRRSGAAGAAVKGVGTIAMELIRMIKRG
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SSIV 246/09 NP      INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQXAMTDQVRESRNPGN
SSIV 45/10 NP       INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
SSIV 132/09 NP      INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
SSIV 199/09 NP      INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
SSIV 18/10 NP       INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
SSIV 206/09 NP      INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
SSIV 294/09 NP      INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
Haseluenne 2003     INDRNFWRGENGRRTRIAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
Mexico 2009         INDRNFWRGENGRRTRVAYERMCNILKGKFQTAAQRAMMDQVRESRNPGN
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SSIV 246/09 NP      AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVXSGYDFEREGYSLVG
SSIV 45/10 NP       AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVASGYDFEREGYSLVG
SSIV 132/09 NP      AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVASGYDFEREGYSLVG
SSIV 199/09 NP      AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVASGYDSEREGYSLVG
SSIV 18/10 NP       AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVASGYDFEREGYSLVG
SSIV 206/09 NP      AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVASGYDFEREGYSLVG
SSIV 294/09 NP      AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVASGYDFEREGYSLVG
Haseluenne 2003     AEIEDLIFLARSALILRGSVAHKSCLPACVYGLIVASGYDFEREGYSLVG
Mexico 2009         AEIEDLIFLARSALILRGSVAHKSCLPACVYGLAVASGHDFEREGYSLVG
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SSIV 246/09 NP      VDPFXLLQNSQVFSLIRPNENXVHKSQLVWMACHSAAFEDLRVSSFIXGT
SSIV 45/10 NP       VDPFRLLQNSQVFSLIRPNENPVHKSQLVWMACHSAAFEDLRVSSFIRGT
SSIV 132/09 NP      VDPFRLLQNSQVFSLIRPNENPVHKSQLVWMACHSAAFEDLRVSSFIRGT
SSIV 199/09 NP      VDPFRLLQNSQVFSLIRPNENPVHKSQLVWMACHSAAFEDLRVSSFIRGT
SSIV 18/10 NP       IDPFRLLQNSQVFSLIRPNENPVHKSQLVWMACHSAAFEDLRVSSFIRGT
SSIV 206/09 NP      IDPFRLLQNSQVFSLIRPNENPVHKSQLVWMACHSAAFEDLRVSSFIRGT
SSIV 294/09 NP      IDPFRLLQNSQVFSLIRPNENPVHKSQLVWMACHSAAFEDLRVSSFIRGT
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Haseluenne 2003     IDPFRLLQNSQVFSLIRPNENPVHKSQLIWMACHSAAFEDLRVSSFIRGT
Mexico 2009         IDPFKLLQNSQVVSLMRPNENPAHKSQLVWMACHSAAFEDLRVSSFIRGK
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SSIV 246/09 NP      KVIPRGQLSTRGVQIASNENMETMDSITLXLRSKXWAIRTRXGGNTNQQR
SSIV 45/10 NP       KVIPRGQLSTRGVQIASNENMETMDSITLELRSKYWAIRTRSGGNTNQQR
SSIV 132/09 NP      KVIPRGQLSTRGVQIASNENMETMDSITLELRSKYWAIRTRSGGNTNQQR
SSIV 199/09 NP      KVIPRGQLSTRGVQIASNENMETMDSITLELRSKYWAIRTRSGGNTNQQR
SSIV 18/10 NP       KVIPRGQLSTRGIQIASNENMETMDSITLELRSKYWAIRTRSGGNTNQQR
SSIV 206/09 NP      KVIPRGQLSTRGIQIASNENMETMDSITLELRSKYWAIRTRSGGNTNQQR
SSIV 294/09 NP      KVIPRGQLSTRGVQIASNENMETMDSITLELRSKYWAIRTRSGGNTNQQR
Haseluenne 2003     KVIPRGQLSTRGVQIASNENMETMDSITLELRSKYWAIRTRSGGNTNQQR
Mexico 2009         KVIPRGKLSTRGVQIASNENVEIMDSNTLELRSRYWAIRTRSGGNTNQQK
                    ******:*****:*******:* *** ** ***: ****** *******:

SSIV 246/09 NP      ASAGQISXQPTFSXQXNLPFERATIMAAFXGNTEGRTSDMRTEIIRMMES
SSIV 45/10 NP       ASAGQISVQPTFSVQRNLPFERATIMAAFTGNTEGRTSDMRTEIIKMMES
SSIV 132/09 NP      ASAGQISVQPTFSVQRNLPFERATIMAAFTGNTEGRTSDMRTEIIRMMES
SSIV 199/09 NP      ASAGQISVQPTFSVQRNLPFERATIMAAFTGNTEGRTSDMRTEIIRMMES
SSIV 18/10 NP       ASAGQISVQPTFSVQRNLPFERATIMAAFTGNTEGRTSDMRTEIIRMMES
SSIV 206/09 NP      ASAGQISVQPTFSVQRNLPFERATIMAAFTGNTEGRTSDMRTEIIRMMES
SSIV 294/09 NP      ASAGQISVQPTFSVQRNLPFERATIMAAFTGNTEGRTSDMRTEIIRMMES
Haseluenne 2003     ASAGQISVQPTFSVQRNLPFERATIMAAFTGNTEGRTSDMRTEIIRMMES
Mexico 2009         ASAGQISVQPTFSVQRNLPFERATVMAAFSGNNEGRTSDMRTEVIRMMES
                    ******* ***** * ********:**** **.**********:*:****

SSIV 246/09 NP      ARPEXVSFQGRGVFELSDEKATXPIVPSFDMSNEGSYXFGDNAEEYDN
SSIV 45/10 NP       ARPEDVSFQGRGVFELSDEKATSPIVPSFDMSNEGSYFFGDNAEDYDS
SSIV 132/09 NP      ARPEDVSFQGRGVFELSDEKATNPIVPSFDMSNEGSYFFGDNAEEYDS
SSIV 199/09 NP      ARPEDVSFQGRGVFELSDEKATNPIVPSFDMSNEGSYFFGDNAEEYDS
SSIV 18/10 NP       ARPEDVSFQGRGVFELSDEKATNPIVPSFDMSNEGSYFFGDNAEEYDS
SSIV 206/09 NP      ARPEDVSFQGRGVFELSDEKATNPIVPSFDMSNEGSYFFGDNAEEYDS
SSIV 294/09 NP      ARPEDVSFQGRGVFELSDEKATNPIVPSFDMNNEGSYFFGDNAEEYDS
Haseluenne 2003     ARPEDVSFQGRGVFELSDEKATNPIVPSFDMSNEGSYFFGDNAEEYDN
Mexico 2009         AKPEDLSFQGRGVFELSDEKATNPIVPSFDMSNEGSYFFGDNAEEYDS
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Legend:
Amino acid sequences of the nucleoprotein. 301 V/I and 363 V/I are highlighted in blue. Positions 366-374 highlighted in red, with the differences in the pandemic strain Mexico 2009 in yellow.

Appendix 5a

Segment 4

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SSIV 18/10 HA       ---ACGCGTGATN--GCAAAANCAGGGGATAATTAATTAAATAAACCAAA
SSIV 206/09 HA      ---ACGCGTGATCA-GCAAAAGCATGGGATAATTAATTAAATAAACCAAA
SSIV 270/09 HA      ---ACGCGTGATCA-GCAAAAGCAGGGGATAATTA----AATAAACCAAA
SSIV 132/09 HA      ---ACNCGTNATCN-NCANAAGCNNGGGATANTTAATTAAANAAACCAAA
SSIV 45/10 HA       ---TCGCGTGATCA-GCAAAAGCAGGGGATAATTA----AATAAACCAAG
Haseluenne 2003     TATTCGTATCAGGGAGCAAAAGCAGGGGATAATTA----AATCAACCAAA
Mexico 2009         -------------------------------------AAAAGCAACAAAA
                                                        **  *** **

SSIV 18/10 HA       ATGAAAGCAGAATTGTTTGTACTATTCTGTGCATTCACTGCACTGAAAGC
SSIV 206/09 HA      ATGAAAGCAGAATTGTTTGTACTATTCTGTGCATTCACTGCACTGAAAGC
SSIV 270/09 HA      ATGAAAGCAGAATTGTTTGTACTATTCTGTGCATTCACTGCACTGAAAGC
SSIV 132/09 HA      ATGAAAGCAGAATTGTTTGNNCTATTNTGNGCNTNCACTGNACTGNNNGC
SSIV 45/10 HA       ATGAAAGCAAAATTGTTTGTACTATTCTGTGCATTCACTGCACTGAGAGC
Haseluenne 2003     ATGGAAGCAAAACTGTTTGTGTTATTCTGTGCATTCACAGCACTAAAAGC
Mexico 2009         ATGAAGGCAATACTAGTAGTTCTGCTATATACATTTGCAACCGCAAATGC
                    *** * ***  * *  * *   *  * *   * *   *          **

SSIV 18/10 HA       TGACACTATTTGTGTAGGCTATCATGCTAACAATTCCACAGACACTGTCG
SSIV 206/09 HA      TGACACTATTTGTGTAGGCTATCATGCTAACAATTCCACAGACACTGTCG
SSIV 270/09 HA      TGACACTATTTGTGTAGGCTATCATGCTAACAATTCCACAGACACTGTCG
SSIV 132/09 HA      TGACACTATTTGTGTNNGCTATNNTGCTAACAATTCCACNGACNCNGTNN
SSIV 45/10 HA       TGACACTATTTGTGTAGGCTATCATGCTAACAATTCCACAGACACCGTCG
Haseluenne 2003     AGACACCATTTGTGTAGGCTATCATGCTAACAATTCCACAGACACTGTCG
Mexico 2009         AGACACATTATGTATAGGTTATCATGCGAACAATTCAACAGACACTGTAG
                    *****  * *** *  * ***  *** ******** ** *** * **

SSIV 18/10 HA       ACACAATACTGGAGAGGAATGTGACTGTTACCCATTCAGTTAATTTGCTA
SSIV 206/09 HA      ACACGATACTGGAGAGGAATGTGACTGTTACCCATTCAGTTAATTTGCTA
SSIV 270/09 HA      ACACAATACTGGAGAGGAATGTGACTGTTACCCATTCAGTTAATTTGCTC
SSIV 132/09 HA      NCACGATACTGGANAGNANTGTGNNTGTTANCCATTCANTTAANTNGCTN
SSIV 45/10 HA       ACACAATACTGGAGAAGAATGTGACTGTTACCCATTCAGTTAATTTGCTA
Haseluenne 2003     ACACAATACTGGAGAAGAATGTGACTGTTACCCATTCAATTAACTTACTA
Mexico 2009         ACACAGTACTAGAAAAGAATGTAACAGTAACACACTCTGTTAACCTTCTA
                    ***  **** ** *  * ***    ** *  ** **  ****    **

SSIV 18/10 HA       GAAAACAGCCACAATGGAAAACTCTGCAGCCTGAATGGAAAGGCCCCATT
SSIV 206/09 HA      GAAAACAGCCACAATGGAAAACTCTGCAGCCTGAATGGAAAGGCCCCATT
SSIV 270/09 HA      GAAAACAGCCACAATGGAAAACTCTGCAGCCTGAATGGAAAGGCCCCATT
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SSIV 132/09 HA     NAAAACAGCCNCAATGGAANACTCTGCAGCCTGAATGGNAAGGCCCNATT
SSIV 45/10 HA      GAAAACAGCCACAATGGAAAACTCTGCAGCCTAAATGAAAAGGCCCCATT
Haseluenne 2003    GAAAACAACCATAATGGAAAACTCTGCAGCCTGAATGGGAAAGCCCCCTT
Mexico 2009        GAAGACAAGCATAACGGGAAACTATGCAAACTAAGAGGGGTAGCCCCATT
                    ** ***  *  ** ** * *** ****  ** *  *     ****  **

SSIV 18/10 HA      ACAACTGGGGAACTGCAACGTAGCAGGATGGATCCTTGGAAACCCAGAAT
SSIV 206/09 HA     ACAACTGGGGAACTGCAACGTAGCAGGATGGATCCTTGGAAACCCAGAAT
SSIV 270/09 HA     ACAACTGGGGAACTGCAACGTAGCAGGATGGATCCTTGGGAACCCAGAAT
SSIV 132/09 HA     ACAACTGGGGAACTGCNNCGTANNNNGATGGATCCNTGGAANCCCNGAAT
SSIV 45/10 HA      ACAACTGGGGAACTGCAACGTAGCAGGATGGATCCTTGGCAACCCAGAAT
Haseluenne 2003    ACAACTGGGGAACTGCAACGTAGCAGGATGGATCCTGGGCAACCCAGAAT
Mexico 2009        GCATTTGGGTAAATGTAACATTGCTGGCTGGATCCTGGGAAATCCAGAGT
                    **  **** ** **   * *     * *******  ** *  ** ** *

SSIV 18/10 HA      GCGACTTGCTGCTCACAGTGAATTCATGGTCTTACATAATAGAGACTTCA
SSIV 206/09 HA     GCGACTTGCTGCTCACAGTGAATTCGTGGTCTTACATAATAGAGACTTCA
SSIV 270/09 HA     GCGACTTGCTGCTCACAGTGAATTCGTGGTCTTACATAATAGAGACTTCA
SSIV 132/09 HA     GNGACNTGCTGCTCACANTNNATTNNNGNTCTTACATAATAGANACTTCN
SSIV 45/10 HA      GCGACTTGCTGCTCACAGTGAATTCGTGGTCTTACATAATAGAGACTTCA
Haseluenne 2003    GTGACTTGCTGCTCACAGTGGATTCGTGGTCTTACATAATAGAGACTTCA
Mexico 2009        GTGAATCAATCTCCACAGCAAGCTCATGGTCCTACATTGTGGAAACATCT
                   * **     *   ****      *   * ** *****  * ** ** **

SSIV 18/10 HA      AATTCAAAGAATGGGGCATGCTATCCTGGAGAATTCGCTGATTATGAAGA
SSIV 206/09 HA     AATTCAAAGAATGGGGCATGCTATCCTGGAGAATTCGCTGATTATGAAGA
SSIV 270/09 HA     AATTCAAAGAATGGGGCATGCTATCCTGGAGAATTCGCTGATTATGAAGA
SSIV 132/09 HA     AATTCAAAGAATGGGGCATGCTATCCTGNAGAATTCGNTGATTATGAAGA
SSIV 45/10 HA      AATTCAAAAAATGGGACATGCTATCCTGGAGAATTCGCTGATTATGAGGA
Haseluenne 2003    AATTCAAAGAATGGAGCATGCTACCCCGGAGAATTCGCTGATTATGAAGA
Mexico 2009        AGTTCAGACAATGGAACGTGTTACCCAGGAGATTTCATCGATTATGAGGA
                   * **** * *****  * ** ** ** * *** ***   ******** **

SSIV 18/10 HA      ATTAAGGGAGCAGCTGGGTACAGTTTCTTCATTTGAAAGATTTGAAATTT
SSIV 206/09 HA     ATTAAGGGAGCAGCTGGGTACAGTTTCTTCATTTGAAAGATTTGAAATTT
SSIV 270/09 HA     ATTAAGGGAGCAGCTGAGTACAGTTTCTTCATTTGAAAGATTTGAAATTT
SSIV 132/09 HA     ATTNAGNNAGCNGNTNGNTNCANNTNCTTCANTNGAAAGATTTGANANTT
SSIV 45/10 HA      ATTGAGGGAGCAGCTGAGTACAGTTTCTTCATTTGAAAGATTTGAAATTT
Haseluenne 2003    ATTAAGGGAGCAGCTGAGTACAGTTTCTTCATTTGAAAGATTTGAAATTT
Mexico 2009        GCTAAGAGAGCAATTGAGCTCAGTGTCATCATTTGAAAGGTTTGAGATAT
                     * **  ***   *     **    * *** * ***** ***** *  *

SSIV 18/10 HA      TCCCAAAAGCAACCTCATGGCCAAATCATGAGACAACCAAAGGTACCACA
SSIV 206/09 HA     TCCCAAAAGCAACCTCATGGCCAAATCATGAGACAACCAAAGGTACCACA
SSIV 270/09 HA     TCCCAAAAGCAACCTCATGGCCAAATCATGAGACAACCAAAGGTACCACA
SSIV 132/09 HA     NCCCAAAAGCAACCTCATGGCCNAATCANGAGACAACNANNGGTACCNCN
SSIV 45/10 HA      TCCCAAAAGCAACCTCATGGCCAAATCATGATACAATCAAAGGTACTACA
Haseluenne 2003    TCCCTAAAGCAACCTCATGGCCAAATCATGATACAACCAGAGGTACCACA
Mexico 2009        TCCCCAAGACAAGTTCATGGCCCAATCATGACTCGAACAAAGGTGTAACG
                    *** **  ***  ******** ***** **  * *  *  ***    *

SSIV 18/10 HA      ATTGCATGCTCCCACTCTGGAGCAAACAGTTTTTATCGGAACTTGCTATG
SSIV 206/09 HA     ATTGCATGCTCCCACTCTGGAGCCAACAGTTTTTATCGGAACTTGCTATG
SSIV 270/09 HA     ATTGCATGCTCCCACTCTGGAGCAAACAGTTTTTATCGGAACTTGCTATG
SSIV 132/09 HA     NTTNCNTGCTCCCACTCTGGAGCCAACANNNTTNATCGNAACTTGCTATN
SSIV 45/10 HA      GTTGCATGCTCCCACTCTGGAGCAAACAGTTTTTATCGGAACTTGCTATG
Haseluenne 2003    ATTTCATGCTCCCACTCTGGAGCCAATAGTTTTTATCGGAACTTGCTATG
Mexico 2009        GCAGCATGTCCTCATGCTGGAGCAAAAAGCTTCTACAAAAATTTAATATG
                       * **  * **  ******* ** *   *  *    ** **  ***

SSIV 18/10 HA      GATAGTAAAAAAGGGAAACTCCTATCCTAAGCTCAGCAAGTCATACACAA
SSIV 206/09 HA     GATAGTAAAAAAGGGAAACTCCTATCCTAAGCTCAGCAAGTCATACACAA
SSIV 270/09 HA     GATAGTAAAAAAGGGAAACTCCTATCCTAAGCTCAGCAAGTCATACACAA
SSIV 132/09 HA     GNTAGTNAAAAAGGNAAACTCCTATCCTAAGCTCANCAAGTCATACACAA
SSIV 45/10 HA      GATAGTAAAAAAGGGAAACTCCTATCCTAAGCTCAGCAAGTCATACACAA
Haseluenne 2003    GATAGTAAAGAAAGGAAACTCCTATCCTAAGCTCAGCAAGTCATACACAA
Mexico 2009        GCTAGTTAAAAAAGGAAATTCATACCCAAAGCTCAGCAAATCCTACATTA
                   * **** ** ** * *** ** ** ** ******* *** ** ****  *

SSIV 18/10 HA      ACAACAAAGGAAAAGAAGTGCTTGTAATCTGGGGAGTTCATCACCCTCCG
SSIV 206/09 HA     ACAACAAAGGAAAAGAAGTGCTTGTAATCTGGGGAGTTCATCACCCTCCG
SSIV 270/09 HA     ACAGCAAAGGAAAAGAAGTGCTTGTAATCTGGGGAGTTCATCACCCTCCG
SSIV 132/09 HA     ACNACANNGGAAAAGAANTGCTTGTNNNCTGNGGAGTTCATCACCCTCCG
SSIV 45/10 HA      ACAACAAAGGAAAAGAAGTGCTTGTAATCTGGGGAGTTCATCACCCTCCG
Haseluenne 2003    ACAACAAAGGAAAAGAAGTGCTTGTAATCTGGGGAGTGCACCACCCTCCG
Mexico 2009        ATGATAAAGGGAAAGAAGTGCTCGTGCTATGGGGCATTCACCATCCATCT
                   *    *  ** ****** * ** **    ** **  * ** ** **  *

SSIV 18/10 HA      ACTAACAGTGATCAACAAACCCTCTACCAGAATAATCACACATATGTTTC
SSIV 206/09 HA     ACTAACAGTGATCAACAAACCCTCTACCAGAATAATCACACATATGTTTC
SSIV 270/09 HA     ACTAACAGTGATCAACAAACCCTCTACCAGAATAATCACACATATGTTTC
SSIV 132/09 HA     ACTAACAGTGATCAACANACCCTCTACCAGANTAATCACACATATGTTTC
SSIV 45/10 HA      ACTGACAGTGATCAACAAACCCTCTACCAGAATAATCACACATATGTTTC
Haseluenne 2003    ACTGACAGTGACCAACAAACCCTTTACCAAAATAATCATACATATGTTTC
Mexico 2009        ACTAGTGCTGACCAACAAAGTCTCTATCAGAATGCAGATGCATATGTTTT
                   ***     *** ***** *  ** ** ** * *    *  *********

SSIV 18/10 HA      AGTTGGATCATCAAAATACTACCAAAGGTTCACACCAGAAATAGTGACCA
SSIV 206/09 HA     AGTTGGATCATCAAAATACTACCAAGGGTTCACACCAGAAATAGTGACCA
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SSIV 270/09 HA     AGTTGGATCATCAAAATACTACCAAAGGTTCACACCAGAAATAGTGACCA
SSIV 132/09 HA     ANTTGGATCATCANAATACTACCNAGGGTTCACACCAGAAATAGTGACCA
SSIV 45/10 HA      AGTTGGATCATCAAAATACTACCAAAGGTTCACACCAKAAATAGTGACCA
Haseluenne 2003    AGTTGGATCATCAAAATACTACCAAAGGTTCACACCAGAAATAGTAACCA
Mexico 2009        TGTGGGGTCATCAAGATACAGCAAGAAGTTCAAGCCGGAAATAGCAATAA
                     * ** ******  ****  *     *****  **  ******  *  *

SSIV 18/10 HA      GACCTAAAGTCAGAGAGCAAGCGGGCAGAATGAATTATTATTGGACACTA
SSIV 206/09 HA     GACCTAAAGTCAGAGAGCAAGCGGGCAGAATGAATTATTATTGGACACTA
SSIV 270/09 HA     GACCTAAAGTCAGAGAGCAAGCAGGCAGAATGAATTATTATTGGACACTA
SSIV 132/09 HA     GACCTAAAGTCAGAGAGCAAGCGGGCAGAATGAATTATTATTGGACACTA
SSIV 45/10 HA      GACCTAAAGTAAGAGAGCAAGCGGGCAGAATGAATTATTATTGGACACTA
Haseluenne 2003    GACCTAAAGTCAGAGGACAAGCAGGCAGAATGAATTATTATTGGACACTG
Mexico 2009        GACCCAAAGTGAGGGATCAAGAAGGGAGAATGAACTATTACTGGACACTA
                   **** ***** ** *  ****  ** ******** ***** ********

SSIV 18/10 HA      TTAGATCAAGGAGACACCATAACCTTTGAAGCCACGGGGAATTTAATAGC
SSIV 206/09 HA     TTAGATCAAGGAGACACCATAACCTTTGAAGCCACGGGGAATTTAATAGC
SSIV 270/09 HA     TTAGATCAAGGAGACACCATAACCTTTGAAGCCACGGGGAATTTAATAGC
SSIV 132/09 HA     TTAGATCAAGGAGACACCATAACCTTTGAAGCCACGGGGAATTTAATAGC
SSIV 45/10 HA      TTAGATCAAGGAGACACCATAACTTTTGAAGCCACGGGGAATTTAATAGC
Haseluenne 2003    TTAGATCAAGGAGACACAATAACTTTTGAAGCTACTGGGAACTTAATAGC
Mexico 2009        GTAGAGCCGGGAGACAAAATAACATTCGAAGCAACTGGAAATCTAGTGGT
                    **** *  *******  ***** ** ***** ** ** **  ** * *

SSIV 18/10 HA      ACCATGGCATGCATTTGCATTGAATAAGGGCTCTAGTTCTGGAATTATGA
SSIV 206/09 HA     ACCATGGCATGCATTTGCATTGAATAAGGGCTCTAGTTCTGGAATTATGA
SSIV 270/09 HA     ACCATGGCATGCATTTGCATTGAATAAGGGCTCTAGTTCTGGAATTATGA
SSIV 132/09 HA     ACCATGGCATGCATTTGCATTGAATAAGGGCTCTAGTTCTGGAATTATGA
SSIV 45/10 HA      ACCATGGCATGCATTTGCATTGAATAAGGGCTCTAGTTCTGGAATTATGA
Haseluenne 2003    ACCGTGGCACGCATTTGCATTGAATAAGGGCCCTAGTTCTGGAATTATGA
Mexico 2009        ACCGAGATATGCATTCGCAATGGAAAGAAATGCTGGATCTGGTATTATCA
                   ***  *  * ***** *** ** * *      ** * ***** ***** *

SSIV 18/10 HA      TGTCGGATGCTCATGTTCACAATTGCACTACAAAGTGCCAAACTCCTCAT
SSIV 206/09 HA     TGTCGGATGCTCATGTTCACAATTGCACTACAAAGTGCCAAACTCCTCAT
SSIV 270/09 HA     TGTCGGATGCTCATGTTCACAATTGCACTACAAAGTGCCAAACTCCTCAT
SSIV 132/09 HA     TGTCGGATGCTCATGTTCACAATTGCACTACAAAGTGCCAAACTCCTCAT
SSIV 45/10 HA      TGTCGGATGCTCTTGTTCACAATTGCACTACAAAGTGCCAAACTCCTCAT
Haseluenne 2003    TATCGGATGCTCATGTTCACAATTGCACTACAAAGTGCCAAACTCCTCAT
Mexico 2009        TTTCAGATACACCAGTCCACGATTGCAATACAACTTGTCAGACACCCAAG
                   * ** *** * *  ** *** ****** *****  ** ** ** **  *

SSIV 18/10 HA      GGGGCTTTGAAAAGCAATCTTCCTTTTCAGAATGTACATCCCATCACTAT
SSIV 206/09 HA     GGGGCTTTGAAAAGCAATCTTCCTTTTCAGAATGTACATCCCATCACTAT
SSIV 270/09 HA     GGGGCTTTGAAAAGCAATCTTCCTTTTCAGAATGTACATCCCATCACTAT
SSIV 132/09 HA     GGGGCTTTGAAAAGCAATCTTCCTTTTCAGAATGTACATCCCATCACTAT
SSIV 45/10 HA      GGGGCTTTGGAAAGCAATCTTCCTTTTCAGAATGTACATCCCATCACTAT
Haseluenne 2003    GGGGCCCTGAAAAGCAATCTTCCTTTTCAGAACGTACATCCCAGCACTAT
Mexico 2009        GGTGCTATAAACACCAGCCTCCCATTTCAGAATATACATCCGATCACAAT
                   ** **  *  * * **  ** ** ********  ******* * *** **

SSIV 18/10 HA      TGGAGAATGCCCCAAATATGTTAAAAGCACCCAACTAAGAATGGCAACAG
SSIV 206/09 HA     TGGAGAATGCCCCAAATATGTTAAAAGCACCCAACTAAGAATGGCAACAG
SSIV 270/09 HA     TGGAGAATGCCCCAAATATGTTAAAAGCACCCAACTAAGAATGGCAACAG
SSIV 132/09 HA     TGGAGAATGCCCCAAATATGTTAAAAGCACCCAACTAAGAATGGCAACAG
SSIV 45/10 HA      TGGAGAATGCCCCAAATATGTTAAAAGCACCCAACTAAGAATGGCAACAG
Haseluenne 2003    TGGAGAATGCCCCAAATATGTTAAAAGCACCCAACTGAGAATGGCAACAG
Mexico 2009        TGGAAAATGTCCAAAATATGTAAAAAGCACAAAATTGAGACTGGCCACAG
                   **** **** ** ******** ********  ** * *** **** ****

SSIV 18/10 HA      GATTAAGGAATATCCCCTCCATTCAATCCAGAGGACTTTTTGGGGCAATT
SSIV 206/09 HA     GATTAAGGAATATCCCCTCCATTCAATCCAGAGGACTTTTTGGGGCAATT
SSIV 270/09 HA     GATTAAGGAATATTCCCTCCATTCAATCCAGAGGACTTTTTGGGGCAATT
SSIV 132/09 HA     GATTAAGGAATATCCCCTCCATTCAATCCAGAGGACTTTTTGGGGCAATT
SSIV 45/10 HA      GATTAAGGAATATCCCCTCCATTCAATCCAGAGGACTTTTTGGGGCAATT
Haseluenne 2003    GATTAAGGAACATCCCCTCAATTCAATCCAGAGGACTTTTTGGGGCAATT
Mexico 2009        GATTGAGGAATGTCCCGTCTATTCAATCGAGAGGCCTATTTGGGGCCATT
                   **** *****  * ** ** ******** ***** ** ******** ***

SSIV 18/10 HA      GCCGGATTCATTGAAGGAGGATGGACAGGAATGATAGATGGATGGTATGG
SSIV 206/09 HA     GCCGGATTCATTGAAGGAGGATGGACAGGAATGATAGATGGATGGTATGG
SSIV 270/09 HA     GCCGGATTCATTGAAGGAGGATGGACAGGAATGATAGATGGATGGTATGG
SSIV 132/09 HA     GCCGGATTCATTGAAGGAGGATGGACAGGAATGATAGATGGATGGTATGG
SSIV 45/10 HA      GCCGGGTTCATTGAAGGAGGATGGACAGGAATGATAGATGGATGGTATGG
Haseluenne 2003    GCAGGATTCATTGAAGGAGGATGGACAGGAATGATAGATGGATGGTATGG
Mexico 2009        GCCGGTTTCATTGAAGGGGGGTGGACAGGGATGGTAGATGGATGGTACGG
                   ** ** *********** ** ******** *** ************* **

SSIV 18/10 HA      ATATCACCATCAAAATGAGCAGGGGTCTGGTTACGCAGCAGATCAGAAAA
SSIV 206/09 HA     ATATCACCATCAAAATGAGCAGGGGTCTGGTTACGCAGCAGATCAGAAAA
SSIV 270/09 HA     ATATCACCATCAAAATGAGCAGGGGTCTGGTTACGCAGCAGATCAGAAAA
SSIV 132/09 HA     ATATCACCATCAAAATGAGCAGGGGTCTGGTTACGCAGCAGATCAGAAAA
SSIV 45/10 HA      ATATCACCATCAAAATGAGCAGGGATCTGGTTACGCAGCAGATCAGAAAA
Haseluenne 2003    ATATCACCATCAAAATGAGCAAGGGTCTGGTTACGCAGCAGATCAGAAAA
Mexico 2009        TTATCACCATCAAAATGAGCAGGGGTCAGGATATGCAGCCGACCTGAAGA
                   ******************** ** ** ** ** ***** ** * *** *

SSIV 18/10 HA      GTACACAAATCGCAATTGATGGGATCAGCAACAAAGTAAACTCAATAATT
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SSIV 206/09 HA     GTACACAAATCGCAATTGATGGGATCAGCAACAAAGTAAACTCAATAATT
SSIV 270/09 HA     GTACACAAATCGCAATTGATGGGATCAGCAACAAAGTAAACTCAGTAATT
SSIV 132/09 HA     GTACACAAATCGCAATTGATGGGATCAGCAACAAAGTAAACTCAATAATT
SSIV 45/10 HA      GTACACAAATCGCAATTGATGGGATCAGCAACAAAGTAAACTCAATAATT
Haseluenne 2003    GCACACAAATCGCAATTGATGGGATCAACAACAAAGTGAACTCAATAATT
Mexico 2009        GCACACAGAATGCCATTGACGAGATTACTAACAAAGTAAATTCTGTTATT
                   * ***** *  ** ***** * *** *  ******** ** **  * ***

SSIV 18/10 HA      GAAAAAATGAATGTTCAATTTACTTCAGTGGGCAAAGAGTTCAACAACCT
SSIV 206/09 HA     GAAAAAATGAATGTTCAATTTACTTCAGTGGGCAAAGAGTTCAACAACCT
SSIV 270/09 HA     GAAAAAATGAATGTTCAATTTACTTCAGTGGGCAAAGAGTTCAACAACCT
SSIV 132/09 HA     GAAAAAATGAATGTTCAATTTACTTCAGTGGGCAAAGAGTTCAACAACCT
SSIV 45/10 HA      GAAAAAATGAATGTTCAATTTACTTCAGTGGGCAAAGAGTTCAATAATCT
Haseluenne 2003    GAAAAAATGAACACTCAATTTACTTCAGTGGGCAAGGAGTTCAATGATCT
Mexico 2009        GAAAAGATGAATACACAGTTCACAGCAGTAGGTAAAGAGTTCAACCACCT
                   ***** *****    ** ** **  **** ** ** ********  * **

SSIV 18/10 HA      AGAGAAAAGGATTGAGAATTTGAATAAAAAGGTCGATGATGGATTTTTGG
SSIV 206/09 HA     AGAGAAAAGGATTGAGAATTTGAATAAAAAGGTCGATGATGGATTTTTGG
SSIV 270/09 HA     AGAGAAAAGGATTGAGAATTTGAATAAAAAGGTCGATGATGGATTTTTGG
SSIV 132/09 HA     AGAGAAAAGGATTGAGAATTTGAATAAAAAGGTCGATGATGGATTTTTGG
SSIV 45/10 HA      AGAGAAAAGGATTGAGAATTTGAATAAAAAGGTCGATGATGGATTTTTGG
Haseluenne 2003    AGAAAAAAGAATTGAGAATTTGAATAAGAAAGTCGATGATGGGTTTTTGG
Mexico 2009        GGAAAAAAGAATAGAGAATTTAAATAAAAAAGTTGATGATGGTTTCCTGG
                    ** ***** ** ******** ***** ** ** ******** **  ***

SSIV 18/10 HA      ATGTATGGACTTATAACGCTGAGTTACTCATCCTGCTCGAAAACGAAAGA
SSIV 206/09 HA     ATGTATGGACTTATAACGCTGAGTTACTCATCCTGCTCGAGAACGAAAGA
SSIV 270/09 HA     ATGTATGGACTTATAACGCTGAGTTACTCATCCTGCTCGAGAACGAAAGA
SSIV 132/09 HA     ATGTATGGACTTATAACGCTGAGTTACTCATCCTGCTCGAGAACGAAAGA
SSIV 45/10 HA      ATGTATGGACTTATAACGCTGAATTACTCATCCTGCTCGAGAACGAAAGA
Haseluenne 2003    ATGTATGGACATATAACGCTGAGTTGCTCATTTTACTCGAGAACGAAAGG
Mexico 2009        ACATTTGGACTTACAATGCCGAACTGTTGGTTCTATTGGAAAATGAAAGA
                   *  * ***** ** ** ** **  *  *  *  *  * ** ** *****

SSIV 18/10 HA      ACCCTAGATTTTCATGACTTTAACGCGAAAAATCTATATGAAAGGGTCAA
SSIV 206/09 HA     ACCCTAGATTTTCATGACTTTAACGTGAAAAATCTATATGAAAGGGTCAA
SSIV 270/09 HA     ACCCTAGATTTTCATGACTTTAACGTGAAAAATCTGTATGAAAGGGTCAA
SSIV 132/09 HA     ACCCTAGATTTTCATGACTTTAACGTGAAAAATCTATATGAAAGGGTCAA
SSIV 45/10 HA      ACCCTAGATTTCCATGACTTTAACGTGAAAAATCTGTATGAAAGGGTCAA
Haseluenne 2003    ACTCTAGATTTCCATGACTTTAACGTAAAAAATTTATATGAAAAGGTCAA
Mexico 2009        ACTTTGGACTACCACGATTCAAATGTGAAGAACTTATATGAAAAGGTAAG
                   **  * ** *  ** ** *  ** *  ** **  * ******* *** *

SSIV 18/10 HA      GTCACAACTGAGAAACAATGCCAAGGAAATCGGTAATGGCTGTTTTGAGT
SSIV 206/09 HA     GTCACAACTGAGAAACAATGCCAAGGAAATCGGTAATGGCTGTTTTGAGT
SSIV 270/09 HA     GACACAACTGAGAAACAATGCCAAGGAAATCGGTAATGGCTGTTTTGAGT
SSIV 132/09 HA     GTCACAACTGAGAAACAATGCCAAGGAAATCGGTAATGGCTGTTTTGAGT
SSIV 45/10 HA      GTCACAACTAAGAAACAATGCCAAGGAGATCGGTAATGGCTGTTTTGAGT
Haseluenne 2003    ATCACAACTGAGGAACAATGCCAAGGAAATCGGAAATGGCTGTTTTGAGT
Mexico 2009        AAGCCAGCTAAAAAACAATGCCAAGGAAATTGGAAACGGCTGCTTTGAAT
                       ** ** *  ************** ** ** ** ***** ***** *

SSIV 18/10 HA      TCTATCACAAATGTGATAATGAATGCATGGAAAGCGTAAAGAATGGCACA
SSIV 206/09 HA     TCTATCACAAATGTGATAATGAATGCATGGAAAGCGTAAAGAATGGCACA
SSIV 270/09 HA     TCTATCACAAATGTGATAATGAATGCATGGAAAGCGTAAAGAATGGCACA
SSIV 132/09 HA     TCTATCACAAATGTGATAATGAATGCATGGAAAGCGTAAAGAATGGCACA
SSIV 45/10 HA      TCTATCACAAATGTGATAATGAATGCATGGAAAGCGTAAAGAATGGCACA
Haseluenne 2003    TCTATCACAAATGTGATAATGAATGCATGGAAAGTGTAAAGAATGGCACA
Mexico 2009        TTTACCACAAATGCGATAACACGTGCATGGAAAGTGTCAAAAATGGGACT
                   * ** ******** *****    *********** ** ** ***** **

SSIV 18/10 HA      TATAATTATCCCAGATATTCAAAAGAATCCAAATTGAATAGAGAGAAAAT
SSIV 206/09 HA     TATAATTATCCCAGATATTCAAAAGAATCCAAATTGAATAGAGAGAAAAT
SSIV 270/09 HA     TATAATTATCCCAGATATTCAAAAGAATCCAAATTGAATAGAGAGAAAAT
SSIV 132/09 HA     TATAATTATCCCAGATATTCAAAAGNATCCNAATTGNATAGAGANNAAAT
SSIV 45/10 HA      TATAATTATTCCAGATATTCAGAAGAATCCAAATTGAATAGAGAGGAAAT
Haseluenne 2003    TATAATTATCCCAAATATTCAGAAGAATCCAAACTGAATAGAGAGAAAAT
Mexico 2009        TATGACTACCCAAAATACTCAGAGGAAGCAAAATTAAACAGAGAAGAAAT
                   *** * **  * * *** *** * * * *  ** *  * *****  ****

SSIV 18/10 HA      AGACGGTGTAAAACTAGAATCAGTGGGGGTTCATCAGATTTTGGCGATCT
SSIV 206/09 HA     AGACGGTGTAAAACTAGAATCAGTGGGGGTTCATCAGATTTTGGCGATCT
SSIV 270/09 HA     AGACGGTGTAAAACTAGAATCAGTGGGGGTTCATCAGATTTTGGCGATCT
SSIV 132/09 HA     AGACGGTGTNAAACTAGAATCAGTGGGGGTTCATCAGATTTNGGCGATNT
SSIV 45/10 HA      AGACGGTGTAAAACTAGAATCAGTGGGAGTTCATCAGATTTTGGCGATCT
Haseluenne 2003    AGACGGTGTGAAACTAGAATCAATGGGAGTTCACCAGATTTTGGCGATCT
Mexico 2009        AGATGGGGTAAAGCTGGAATCAACAAGGATTTACCAGATTTTGGCGATCT
                   *** ** ** ** ** ******    *  ** * ******* ****** *

SSIV 18/10 HA      ACTCCACAGTCGCCAGTTCCCTGGTATTGCTAGTCTCCCTGGGGGCAATC
SSIV 206/09 HA     ACTCCACAGTCGCCAGTTCCCTGGTATTGCTAGTCTCCCTGGGGGCAATC
SSIV 270/09 HA     ACTCCACAGTCGCCAGTTCCCTGGTATTGCTAGTCTCCCTGGGGGCAATC
SSIV 132/09 HA     ACTCCACAGTCGCCAGTTCCCTGGTATTGCTAGTCNCCCNGGGGGCAATC
SSIV 45/10 HA      ACTCCACAGTCGCCAGTTCCCTGGTATTGCTAGTCTCCCTGGGGGCAATC
Haseluenne 2003    ACTCCACAGTCGCCAGTTCCCTGGTCTTGTTGGTCTCCCTGGGGGCAATC
Mexico 2009        ATTCAACTGTCGCCAGTTCATTGGTACTGGTAGTCTCCCTGGGGGCAATC
                   * ** ** ***********  ****  ** * *** *** **********
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SSIV 18/10 HA      AGCTTCTGGATGTGCTCTAATGGGTCATTGCAATGCAGAATATGCATTTA
SSIV 206/09 HA     AGCTTCTGGATGTGCTCTAATGGGTCATTGCAATGCAGAATATGCATTTA
SSIV 270/09 HA     AGCTTCTGGATGTGCTCTAATGGGTCATTGCAATGCAGAATATGCATTTA
SSIV 132/09 HA     AGNTTNTGGATGTGCTNTAATGGGTCATTGCAANNCAGAATATGCATTTN
SSIV 45/10 HA      AGCTTCTGGATGTGCTCTAATGGGTCATTGCAATGCAGAATATGCATTTA
Haseluenne 2003    AGCTTCTGGATGTGTTCTAATGGGTCATTGCAATGCAGAATATGCATTTA
Mexico 2009        AGTTTCTGGATGTGCTCTAATGGGTCTCTACAGTGTAGAATATGTATTTA
                   ** ** ******** * *********  * **    ******** ****

SSIV 18/10 HA      AGACTTGAATTTCT-AAGTGTACGGAAAAA-CACCCTTGTTTCTACTGAT
SSIV 206/09 HA     AGACTTGAATTTCT-AAGTGTACGGAAAAA-CACCCTTGTTTCTACTGAT
SSIV 270/09 HA     AGACTTGAATTTCG-AAGTGTACGGAAAAA-CACCCTTGTTTCTACTGAT
SSIV 132/09 HA     AGACTTGAATTTNT-AAGTGTACGGAANAA-CACCCNTGTTTNTANTGNN
SSIV 45/10 HA      AGACTTGAATTTCG-AAGTGTACGGAAAAA-CACCCTTGTTTCTACTGAT
Haseluenne 2003    AGACTTGGATTTCA-AAGTGTGCGGAAAAAACACCCTTGTTTCTACTAAT
Mexico 2009        ACATTAGGATTTCAGAAGCAT----------------------------
                   * * * * ****   ***  *

SSIV 18/10 HA      CACGCGT--------
SSIV 206/09 HA     CACGCGT--------
SSIV 270/09 HA     CACGCGA--------
SSIV 132/09 HA     CANGCGT--------
SSIV 45/10 HA      CACGCGT--------
Haseluenne 2003    -ACGAGACGATATAC
Mexico 2009        ---------------
```

Legend:
Nucleotide sequences of segment 4. Highlighted in blue: mutations typical for Swiss strains, causing K39R and S123G. Highlighted in red: sequence affecting the receptor-binding capacity.

Appendix 5b

Haemagglutinin

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SSIV 18/10 HA      MKAELFVLFCAFTALKADTICVGYHANNSTDTVDTILERNVTVTHSVNLL
SSIV 206/09 HA     MKAELFVLFCAFTALKADTICVGYHANNSTDTVDTILERNVTVTHSVNLL
SSIV 270/09 HA     MKAELFVLFCAFTALKADTICVGYHANNSTDTVDTILERNVTVTHSVNLL
SSIV 132/09 HA     MKAELFXLXXAXTXLXADTICVXYXANNSTDXVXTILXXVXVXHSXXXL
SSIV 45/10 HA      MKAKLFVLFCAFTALRADTICVGYHANNSTDTVDTILEKNVTVTHSVNLL
Haseluenne 2003    MEAKLFVLFCAFTALKADTICVGYHANNSTDTVDTILEKNVTVTHSINLL
Mexico 2009        MKAILVVLLYTFATANADTLCIGYHANNSTDTVDTVLEKNVTVTHSVNLL
                   *:* *. *  : :   ***:*: * ****** * *:*   * * **   *

SSIV 18/10 HA      ENSHNGKLCSLNGKAPLQLGNCNVAGWILGNPECDLLLTVNSWSYIIETS
SSIV 206/09 HA     ENSHNGKLCSLNGKAPLQLGNCNVAGWILGNPECDLLLTVNSWSYIIETS
SSIV 270/09 HA     ENSHNGKLCSLNGKAPLQLGNCNVAGWILGNPECDLLLTVNSWSYIIETS
SSIV 132/09 HA     XNSXNGXLCSLNGKAXLQLGNCXVXXWIXGXPEXDXLLTXXXXSYIIXTS
SSIV 45/10 HA      ENSHNGKLCSLNEKAPLQLGNCNVAGWILGNPECDLLLTVNSWSYIIETS
Haseluenne 2003    ENNHNGKLCSLNGKAPLQLGNCNVAGWILGNPECDLLLTVDSWSYIIETS
Mexico 2009        EDKHNGKLCKLRGVAPLHLGKCNIAGWILGNPECESISTASSWSYIVETS
                   :. ** **.*.  * *:**:* :  ** * ** : : *    ***: **

SSIV 18/10 HA      NSKNGACYPGEFADYEELREQLGTVSSFERFEIFPKATSWPNHETTKGTT
SSIV 206/09 HA     NSKNGACYPGEFADYEELREQLGTVSSFERFEIFPKATSWPNHETTKGTT
SSIV 270/09 HA     NSKNGACYPGEFADYEELREQLSTVSSFERFEIFPKATSWPNHETTKGTT
SSIV 132/09 HA     NSKNGACYPXEFXDYEEXXXXXXXXXXSXERFXXXPKATSWPNXETTXGTX
SSIV 45/10 HA      NSKNGTCYPGEFADYEELREQLSTVSSFERFEIFPKATSWPNHDTIKGTT
Haseluenne 2003    NSKNGACYPGEFADYEELREQLSTVSSFERFEIFPKATSWPNHDTTRGTT
Mexico 2009        SSDNGTCYPGDFIDYEELREQLSSVSSFERFEIFPKTSSWPNHDSNKGVT
                   *.**:*** :* ****         * ***   **::**** ::  *.

SSIV 18/10 HA      IACSHSGANSFYRNLLWIVKKGNSYPKLSKSYTNNKGKEVLVIWGVHHPP
SSIV 206/09 HA     IACSHSGANSFYRNLLWIVKKGNSYPKLSKSYTNNKGKEVLVIWGVHHPP
SSIV 270/09 HA     IACSHSGANSFYRNLLWIVKKGNSYPKLSKSYTNSKGKEVLVIWGVHHPP
SSIV 132/09 HA     XXCSHSGANXXXRNLLXXVKKXNSYPKLXKSYTNXXGKEXLVXXGVHHPP
SSIV 45/10 HA      VACSHSGANSFYRNLLWIVKKGNSYPKLSKSYTNNKGKEVLVIWGVHHPP
Haseluenne 2003    ISCSHSGANSFYRNLLWIVKKGNSYPKLSKSYTNNKGKEVLVIWGVHHPP
Mexico 2009        AACPHAGAKSFYKNLIWLVKKGNSYPKLSKSYINDKGKEVLVLWGIHHPS
                   *.*:**:   :**:  *** ****** *** *  *** **  *:***.

SSIV 18/10 HA      TNSDQQTLYQNNHTYVSVGSSKYYQRFTPEIVTRPKVREQAGRMNYYWTL
SSIV 206/09 HA     TNSDQQTLYQNNHTYVSVGSSKYYQGFTPEIVTRPKVREQAGRMNYYWTL
SSIV 270/09 HA     TNSDQQTLYQNNHTYVSVGSSKYYQRFTPEIVTRPKVREQAGRMNYYWTL
SSIV 132/09 HA     TNSDQXTLYQXNHTYVSXGSSXYYXGFTPEIVTRPKVREQAGRMNYYWTL
SSIV 45/10 HA      TDSDQQTLYQNNHTYVSVGSSKYYQRFTPXIVTRPKVREQAGRMNYYWTL
Haseluenne 2003    TDSDQQTLYQNNHTYVSVGSSKYYQRFTPEIVTRPKVRGQAGRMNYYWTL
Mexico 2009        TSADQQSLYQNADAYVFVGSSRYSKKFKPEIAIRPKVRDQEGRMNYYWTL
                   *.:** :***  .:**  *** *   *.* *. ***** * *********

SSIV 18/10 HA      LDQGDTITFEATGNLIAPWHAFALNKGSSSGIMMSDAHVHNCTTKCQTPH
SSIV 206/09 HA     LDQGDTITFEATGNLIAPWHAFALNKGSSSGIMMSDAHVHNCTTKCQTPH
SSIV 270/09 HA     LDQGDTITFEATGNLIAPWHAFALNKGSSSGIMMSDAHVHNCTTKCQTPH
SSIV 132/09 HA     LDQGDTITFEATGNLIAPWHAFALNKGSSSGIMMSDAHVHNCTTKCQTPH
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SSIV 45/10 HA     LDQGDTITFEATGNLIAPWHAFALNKGSSSGIMMSDALVHNCTTKCQTPH
Haseluenne 2003   LDQGDTITFEATGNLIAPWHAFALNKGPSSGIMISDAHVHNCTTKCQTPH
Mexico 2009       VEPGDKITFEATGNLVVPRYAFAMERNAGSGIIISDTPVHDCNTTCQTPK
                  :: **.*********:.* :***:::...***::**: **:*.*.****:

SSIV 18/10 HA     GALKSNLPFQNVHPITIGECPKYVKSTQLRMATGLRNIPSIQSRGLFGAI
SSIV 206/09 HA    GALKSNLPFQNVHPITIGECPKYVKSTQLRMATGLRNIPSIQSRGLFGAI
SSIV 270/09 HA    GALKSNLPFQNVHPITIGECPKYVKSTQLRMATGLRNIPSIQSRGLFGAI
SSIV 132/09 HA    GALKSNLPFQNVHPITIGECPKYVKSTQLRMATGLRNIPSIQSRGLFGAI
SSIV 45/10 HA     GALESNLPFQNVHPITIGECPKYVKSTQLRMATGLRNIPSIQSRGLFGAI
Haseluenne 2003   GALKSNLPFQNVHPSTIGECPKYVKSTQLRMATGLRNIPSIQSRGLFGAI
Mexico 2009       GAINTSLPFQNIHPITIGKCPKYVKSTKLRLATGLRNVPSIQSRGLFGAI
                  **:::.*****:** ***:********:**:******:************

SSIV 18/10 HA     AGFIEGGWTGMIDGWYGYHHQNEQGSGYAADQKSTQIAIDGISNKVNSII
SSIV 206/09 HA    AGFIEGGWTGMIDGWYGYHHQNEQGSGYAADQKSTQIAIDGISNKVNSII
SSIV 270/09 HA    AGFIEGGWTGMIDGWYGYHHQNEQGSGYAADQKSTQIAIDGISNKVNSVI
SSIV 132/09 HA    AGFIEGGWTGMIDGWYGYHHQNEQGSGYAADQKSTQIAIDGISNKVNSII
SSIV 45/10 HA     AGFIEGGWTGMIDGWYGYHHQNEQGSGYAADQKSTQIAIDGISNKVNSII
Haseluenne 2003   AGFIEGGWTGMIDGWYGYHHQNEQGSGYAADQKSTQIAIDGINNKVNSII
Mexico 2009       AGFIEGGWTGMVDGWYGYHHQNEQGSGYAADLKSTQNAIDEITNKVNSVI
                  ***********:****************** **** *** *.*****:*

SSIV 18/10 HA     EKMNVQFTSVGKEFNNLEKRIENLNKKVDDGFLDVWTYNAELLILLENER
SSIV 206/09 HA    EKMNVQFTSVGKEFNNLEKRIENLNKKVDDGFLDVWTYNAELLILLENER
SSIV 270/09 HA    EKMNVQFTSVGKEFNNLEKRIENLNKKVDDGFLDVWTYNAELLILLENER
SSIV 132/09 HA    EKMNVQFTSVGKEFNNLEKRIENLNKKVDDGFLDVWTYNAELLILLENER
SSIV 45/10 HA     EKMNVQFTSVGKEFNNLEKRIENLNKKVDDGFLDVWTYNAELLILLENER
Haseluenne 2003   EKMNTQFTSVGKEFNDLEKRIENLNKKVDDGFLDVWTYNAELLILLENER
Mexico 2009       EKMNTQFTAVGKEFNHLEKRIENLNKKVDDGFLDIWTYNAELLVLLENER
                  ****.***:******.******************:********:******

SSIV 18/10 HA     TLDFHDFNAKNLYERVKSQLRNNAKEIGNGCFEFYHKCDNECMESVKNGT
SSIV 206/09 HA    TLDFHDFNVKNLYERVKSQLRNNAKEIGNGCFEFYHKCDNECMESVKNGT
SSIV 270/09 HA    TLDFHDFNVKNLYERVKTQLRNNAKEIGNGCFEFYHKCDNECMESVKNGT
SSIV 132/09 HA    TLDFHDFNVKNLYERVKSQLRNNAKEIGNGCFEFYHKCDNECMESVKNGT
SSIV 45/10 HA     TLDFHDFNVKNLYERVKSQLRNNAKEIGNGCFEFYHKCDNECMESVKNGT
Haseluenne 2003   TLDFHDFNVKNLYEKVKSQLRNNAKEIGNGCFEFYHKCDNECMESVKNGT
Mexico 2009       TLDYHDSNVKNLYEKVRSQLKNNAKEIGNGCFEFYHKCDNTCMESVKNGT
                  ***:** *.*****:*::**:****************** *********

SSIV 18/10 HA     YNYPRYSKESKLNREKIDGVKLESVGVHQILAIYSTVASSLVLLVSLGAI
SSIV 206/09 HA    YNYPRYSKESKLNREKIDGVKLESVGVHQILAIYSTVASSLVLLVSLGAI
SSIV 270/09 HA    YNYPRYSKESKLNREKIDGVKLESVGVHQILAIYSTVASSLVLLVSLGAI
SSIV 132/09 HA    YNYPRYSKXSXLXRXXIDGVKLESVGVHQIXAXYSTVASSLVLLVXXGAI
SSIV 45/10 HA     YNYSRYSEESKLNREEIDGVKLESVGVHQILAIYSTVASSLVLLVSLGAI
Haseluenne 2003   YNYPKYSEESKLNREKIDGVKLESMGVHQILAIYSTVASSLVLLVSLGAI
Mexico 2009       YDYPKYSEEAKLNREEIDGVKLESTRIYQILAIYSTVASSLVLVVSLGAI
                  *:*.:**: : * *  ********  ::** * **********:*  ***

SSIV 18/10 HA     SFWMCSNGSLQCRICI------------------------------
SSIV 206/09 HA    SFWMCSNGSLQCRICI------------------------------
SSIV 270/09 HA    SFWMCSNGSLQCRICI------------------------------
SSIV 132/09 HA    XXWMCXNGSLQXRICIXDLNX-------------------------
SSIV 45/10 HA     SFWMCSNGSLQCRICI-DLNFEVYGKTPLFLLITRKGEFQHTGGRY
Haseluenne 2003   SFWMCSNGSLQCRICI------------------------------
Mexico 2009       SFWMCSNGSLQCRICI------------------------------
                  *** ***** ****
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Legend:
Amino acid sequences of Haemagglutinin. Highlighted in blue K39R and S123G, exclusive for Swiss isolates. Position 190, highlighted in red: Aspartate, determining receptor-binding properties.

Appendix 6a

Segment 3

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SSIV 294/09 PA      GGGCGAATTGGGCCCTCTAGATGCATGCTCGAGCGGCCGCCAGTGTGATG
SSIV 18/10 PA       GGGCGAATTGGGCCCTCTAGATGCATGCTCGAGCGGCCGCCAGTGTGATG
SSIV 247/09 PA      GGGCGAATTGGGCCCTCTAGATGCATGCTCGAGCGGCCGCCAGTGTGATG
Haseluenne 2003     ---------------------------------------------------
Mexico 2009         ---------------------------------------------------


SSIV 294/09 PA      GATATCTGCAGAATTCGCCCTTAATTCGTCTCAGGGAGCGAAAGCAGGTA
SSIV 18/10 PA       GATATCTGCAGAATTCGCCCTTTATTCGTCTCAGGGAGCGAAAGCAGGTA
SSIV 247/09 PA      GATATCTGCAGAATTCGCCCTTTATTCGTCTCAGGGAGCGAAAGCAGGTA
Haseluenne 2003     ----------------------------------AGCGAAAGCAGGTA
Mexico 2009         ---------------------------------------------------


SSIV 294/09 PA      CTGATTCAAAATGGAAGACTTTGTGCGACAGTGCTTCAATCCAATGATCG
SSIV 18/10 PA       CTGATTCAAAATGGAAGACTTTGTGCGACAGTGCTTCAATCCAATGATCG
SSIV 247/09 PA      CTGATTCAAAATGGAAGACTTTGTGCGACAATGCTTCAATCCAATGATCG
Haseluenne 2003     CTGATTCAAAATGGAAGACTTTGTGCGGCAATGCTTCAATCCAATGATCG
Mexico 2009         ----TCCAAAATGGAAGACTTTGTGCGACAATGCTTCAATCCAATGATCG
                        * ********************** ** ******************
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SSIV 294/09 PA      TCGAGCTTGCGGAAAAGATAATGAAAGAACATGGAGAAGACCCGAAAATT
SSIV 18/10 PA       TCGAGCTTGCGGAAAAGACAATGAAAGAACATGGAGAAGACCCGAAAATT
SSIV 247/09 PA      TCGAGCTTGCGGAAAAGACAATGAAAGAACATGGAGAAGACCCGAAAATT
Haseluenne 2003     TCGAGCTTGCGGAAAAACCAATGAAAGAACATGGGGAAGACCCGAAAATT
Mexico 2009         TCGAGCTTGCGGAAAAGGCAATGAAAGAATATGGGGAAGATCCGAAAATC
                    ****************   ********** **** ***** ********

SSIV 294/09 PA      GAAACAAACAAATTTGCTGCAATATGCACACACATGGAAGTCTGTTTCAT
SSIV 18/10 PA       GAAACAAACAAATTTGCTGCAATATGCACACACATGGAAGTTTGTTTCAT
SSIV 247/09 PA      GAAATAAACAAATTTGCTGCAATATGCACACACATGGAAGTCTGTTTCAT
Haseluenne 2003     GAAACAAACAAATTTGCTGCAATATGTACACACCTGGAAGTCTGTTTCAT
Mexico 2009         GAAACTAACAAGTTTGCTGCAATATGCACACATTTGGAAGTTTGTTTCAT
                    ****  ***** ************** *****  ******* ********

SSIV 294/09 PA      GTATTCTGATTTCCACTTCATCGACGAGCGAGGTGAATCAACAATAGTAG
SSIV 18/10 PA       GTATTCTGATTTCCACTCCATCGACGAGCGTGGTGAATCAACAATAGTAG
SSIV 247/09 PA      GTATTCTGATTTTCACTTCATCGACGAGCGAGGTGAATCGACAATAGTAG
Haseluenne 2003     GTATTCAGACTTCCACTTCATTGATGAACGAGGTGAATCAATAGTTGTAG
Mexico 2009         GTATTCGGATTTCCATTTCATCGACGAACGGGGTGAATCAATAATTGTAG
                    ****** ** ** ** * *** ** ** ** ******** * * * ****

SSIV 294/09 PA      AATCCAGTGATCCAAATGCACTCCTAAAACACAGATTTGAAATAATTGAG
SSIV 18/10 PA       AATCCAGTGATCCAAATGCACTCCTAAAACACAGATTTGAAATAATTGAG
SSIV 247/09 PA      AATCCAGTGATCCAAATGCACTCCTAAAACACAGATTTGAAATAATTGAG
Haseluenne 2003     AATCTGGTGATCCAAATGCGCTCCTAAAACACAGATTTGAAATAATTGAG
Mexico 2009         AATCTGGTGACCCGAATGCACTATTGAAGCACCGATTTGAGATAATTGAA
                    ****  **** ** ***** **  * ** *** ******* ********

SSIV 294/09 PA      GGAAGAGATCGCACAGTGGCTTGGACAGTGGTAAACAGCATCTGCAATAC
SSIV 18/10 PA       GGAAGAGATCGCACAGTGGCTTGGACAGTGGTAAACAGCATCTGCAATAC
SSIV 247/09 PA      GGAAGAGATCGCACAGTGGCTTGGACAGTGGTAAACAGCATCTGCAATAC
Haseluenne 2003     GGAAGGGATCGCACAATGGCCTGGACAGTGGTGAACAGCATCTGCAATAC
Mexico 2009         GGAAGAGACCGAATCATGGCCTGGACAGTGGTGAACAGTATATGTAACAC
                    ***** ** ** *   **** *********** ***** ** ** ** **

SSIV 294/09 PA      TACAGGGGTTGAGAAGCCCAAGTTCCTTCCGGATCTATATGACTATAAGG
SSIV 18/10 PA       TACAGGGGTTGAGAAGCCCAAGTTCCTTCCGGATCTATATGACTATAAGG
SSIV 247/09 PA      TACAGGAGTTGAGAAGCCCAAGTTCCTTCCGGATCTATATGACTATAAGG
Haseluenne 2003     TACAGAGGTTGAAAAGCCCAAGTTTCTCCCAGATCTGTATGACTATAAGG
Mexico 2009         AACAGGGGTAGAGAAGCCTAAATTTCTTCCTGATTTGTATGATTACAAAG
                     ****  ** ** ***** ** ** ** ** *** * ***** ** ** *

SSIV 294/09 PA      AGAACCGGTTCATTGAAATCGGTGTAACAAGAAGGGAAGTTCACATATAC
SSIV 18/10 PA       AGAACCGGTTCATTGAAATCGGTGTAACAAGAAGGGGAGTTCACATATAC
SSIV 247/09 PA      AGAACCGGTTCATTGAAATCGGTGTAACAAGAAGGGAAGTTCACATATAC
Haseluenne 2003     AGAACCGATTCATTGAAATTGGTGTAACAAGAAGAGAGGTCCACATATAC
Mexico 2009         AGAACCGGTTCATTGAAATTGGAGTAACACGGAGGGAAGTCCACATATAT
                    ******* *********** ** ****** * ** *  ** ********

SSIV 294/09 PA      TACCTGGAAAAAGCCAACAAAATAAAATCAGAGAAGACACACATCCACAT
SSIV 18/10 PA       TACCTGGAAAAAGCCAACAAAATAAAATCAGAGAAGACACACATCCACAT
SSIV 247/09 PA      TACCTGGAAAAAGCCAACAAAATAAAATCGGAGAAGACACACATCCACAT
Haseluenne 2003     TATCTAGAAAAAGCAAACAAGATAAAATCAGAGAAGACACACATCCACAT
Mexico 2009         TACCTAGAGAAAGCCAACAAAATAAAATCTGAGAAGACACACATTCACAT
                    ** ** ** ***** ***** ******** ************** *****

SSIV 294/09 PA      TTTCTCGTTCACTGGAGAAGAGATGGCCACTAAGGCAGATTACACTCTTG
SSIV 18/10 PA       TTTCTCGTTCACTGGAGAAGAAATGGCCACTAAGGCAGATTACACTCTTG
SSIV 247/09 PA      TTTCTCGTTCACTGGAGAAGAAATGGCCACTAAGGCAGATTACACTCTTG
Haseluenne 2003     TTTCTCGTTCACTGGGGAAGAAATGGCCACTAAGGCAGATTACACTCTTG
Mexico 2009         CTTTTCATTCACTGGAGAGGAGATGGCCACCAAAGCGGACTACACCCTTG
                     ** ** ******** ** ** ******** ** ** ** ***** ****

SSIV 294/09 PA      ACGAGGAAAGCAGAGCAAGAATCAAAACTAGATTATTCACCATCAGGCAG
SSIV 18/10 PA       ACGAGGAAAGCAGAGCAAGAATCAAAACAAGACTATTCACCATCAGGCAG
SSIV 247/09 PA      ACGAGGAGAGCAGAGCAAGAATCAAAACTAGACTATTCACCATCAGGCAG
Haseluenne 2003     ACGAGGAGAGCAGAGCAAGAATCAAGACCAGACTATTCACCATCAGGCAG
Mexico 2009         ACGAAGAGAGCAGGGCAAGAATCAAAACTAGGCTTTTCACTATAAGACAA
                    **** ** ***** ********** ** **  * ***** ** ** **

SSIV 294/09 PA      GAAATGGCCATTAGGGCCCTATGGGACTCCTTTCGTCAGTCCGAAAGAGG
SSIV 18/10 PA       GAAATGGCCATTAGGGCCCTATGGGACTCCTTTCGTCAGTCCGAAAGAGG
SSIV 247/09 PA      GAAATGGCCATTAGGGCCCTATGGGACTCCTTTCGTCAGTCCGAAAGAGG
Haseluenne 2003     GAATTGGCCATTAGGGGCCTATGGGACTCCTTTCGTCAGTCCGAAAGAGG
Mexico 2009         GAAATGGCCAGTAGGAGTCTATGGGATTCCTTTCGTCAGTCCGAAAGAGG
                    *** ****** ****   ******** ************************

SSIV 294/09 PA      CGAAGAGACAATTGAAGAAAGGTTTGAAATCAAGGGAACCATGCATAGAC
SSIV 18/10 PA       CGAAGAGACAATTGAAGAAAGGTTTGAAATCAAAGGAACCATGCATAGGC
SSIV 247/09 PA      CGAAGAGACAATTGAGGAAAGGTTTGAAATCAAGGGAACCATGCATAGGC
Haseluenne 2003     CGAAGAGACAATTGAAGAAAGGTTTGAAATCAAGGGGACCATGCGTAGGC
Mexico 2009         CGAAGAGACAATTGAAGAAAAATTTGAGATTACAGGAACTATGCGCAAGC
                    *************** ****  ***** ** *  ** ** ****  *  *

SSIV 294/09 PA      TTGCCAACCAAAGTCTCCCGCCGAACTTCTCCAGCCTTGAAAACTTCAGA
SSIV 18/10 PA       TTGCCAACCAAAGTCTCCCACCGAACTTCTCCAGCATTGAAAACTTTAGA
SSIV 247/09 PA      TTGCCAACCAAAGTCTCCCGCCAAACTTCTCCAGCCTTGAAAACTTTAGA
Haseluenne 2003     TTGCCAACCAAAGTCTCCCACCGAACTTCTCCAGTCTTGAAAACTTTAGA
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Mexico 2009          TTGCCGACCAAAGTCTCCCACCGAACTTCTCCAGCCTTGAAAACTTTAGA
                     ***** ************* ** ***********  ********** ***

SSIV 294/09 PA       GCCTATGTGGATGGATTCGAACCGAACGGCTGCATTGAGGGCAAGCTTTC
SSIV 18/10 PA        GCCTATGTGGATGGATTCGAACCGAACGGCTGCATTGAGGGCAAGCTTTC
SSIV 247/09 PA       GCCTATGTGGATGGATTCGAACCGAACGGCTGCATTGAGGGCAAGCTTTC
Haseluenne 2003      GCCTATGTGGATGGATTCGAACCGAACAGCTGCATTGAGAGCAAGCTTTC
Mexico 2009          GCCTATGTAGATGGATTCGAGCCGAACGGCTGCATTGAGGGCAAGCTTTC
                     ******** *********** ****** *********** **********

SSIV 294/09 PA       TCAAATGTCCAAAGAAGTGAATGCCAGGATTGAACCATTCTTGAGGGGAA
SSIV 18/10 PA        TCAAATGTCCAAAGAAGTGAATGCCAGGATTGAACCATTCTTGAGGGGAA
SSIV 247/09 PA       TCAAATGTCCAAAGAAGTGAATGCCAGGATTGAACCATTCTTGAGGGGAA
Haseluenne 2003      TCAAATGTCCAAAGAAGTGAATGCCAGGATTGACCCATTCTTGAGGGAAA
Mexico 2009          CCAAATGTCAAAAGAAGTGAACGCCAAAATTGAACCATTCTTGAGGACGA
                      ******** *********** ****  ***** ************   *

SSIV 294/09 PA       CACCACGCCCTCTCAGGTTACCTGATGGACCTCTCTGTTCTCAACGGTCA
SSIV 18/10 PA        CACCACGCCCTCTCAGGTTACCTGATGGACCTCCCTGTTCTCAACGGTCA
SSIV 247/09 PA       CACCACGCCCTCTCAGATTACCTGATGGACCTCCCTGTTCTCAACGGTCA
Haseluenne 2003      CGCCACGCCCTCTCAGATTACCTGATGGACCTCCCTGTTCCCAACGGTCA
Mexico 2009          CACCACGCCCCCTCAGATTGCCTGATGGGCCTCTTTGCCATCAGCGGTCA
                     * ******** ***** ** ******** ****  **    ** ******

SSIV 294/09 PA       AAATTCCTTCTGATGGACGCCTTAAAATTGAGCATTGAGGATCCTAGTCA
SSIV 18/10 PA        AAATTCCTGCTGATGGACGCCTTAAAATTGAGCATTGAGGATCCTAGTCA
SSIV 247/09 PA       AAGTTCCTGCTGATGGACGCCTTGAAATTGAGCATTGAGGATCCTAGTCA
Haseluenne 2003      AAATTCCTGCTTATGGATGCCTTAAAATTAAGCATTGAGGATCCGAGTCA
Mexico 2009          AAGTTCCTGCTGATGGATGCTCTGAAATTAAGTATTGAAGACCCGAGTCA
                     ** ***** ** ***** **  * ***** ** ***** ** ** *****

SSIV 294/09 PA       TGAGGGGGAGGGGATACCGCTATATGATGCAATCAAGTGCATGAAGACAT
SSIV 18/10 PA        TGAGGGGGAGGGGATACCGCTATATGATGCAATCAAGTGCATGAAGACAT
SSIV 247/09 PA       TGAGGGGGAGGGGATACCGCTATATGATGCACTCAAGTGCATGAAGACAT
Haseluenne 2003      TGAGGGGGAGGGAATACCGCTATATGATGCAATCAAGTGCATGAAGACAT
Mexico 2009          CGAGGGGGAGGGAATACCACTATATGATGCAATCAAATGCATGAAGACAT
                      *********** ***** ************ **** **************

SSIV 294/09 PA       TTTTCGGATGGAAAGAGCCCCACATTGTCAAGCCACATGAGAGAGGCATA
SSIV 18/10 PA        TTTTCGGATGGAAAGAGCCCCACATTGTCAAGCCACATGAGAAAGGCATA
SSIV 247/09 PA       TTTTCGGATGGAAAGAGCCCCACATTGTCAAGCCGCATGAGAAAGGCATA
Haseluenne 2003      TTTTCGGGTGGAAGGAACCCAATATTGTCAAACCACATGAGAAAGGCATA
Mexico 2009          TCTTTGGCTGGAAAGAGCCTAACATAGTCAAACCACATGAGAAAGGCATA
                     * ** ** ***** ** **  * ** ***** ** ******* *******

SSIV 294/09 PA       AATCCAAACTATCTCCTGGCTTGGAAACAAATATTAGCTGAACTACAGGA
SSIV 18/10 PA        AATCCAAACTATCTCCTGGCTTGGAAACAAATACTAGCTGAACTACAGGA
SSIV 247/09 PA       AACCCAAACTATCTCCTGGCTTGGAAACAAATACTAGCAGAACTACAGGA
Haseluenne 2003      AACCCCAATTATCTCCTAGCTTGGAAACAGGTGCTAGCAGAGATACAGGA
Mexico 2009          AATCCCAATTACCTCATGGCTTGGAAGCAGGTGCTAGCAGAGCTACAGGA
                     ** ** ** ** *** * ******** **  *  **** **  *******

SSIV 294/09 PA       CATTGAAAATGAGGAGAAAATCCCCAAAACTAAAAACATGAAAAAAACAA
SSIV 18/10 PA        CATTGAAAATGAGGAGAAAATCCCCAAAACTAAAAACATGAAAAAAACAA
SSIV 247/09 PA       CATTGAGAATGAGGAGAAAATCCCCAAAACTAAAAACATGAAAAAAACAA
Haseluenne 2003      CATTGAAAATGAAGAGAAAATTCCAAAGACAAAAAACATGAAGAAAACAA
Mexico 2009          CATTGAAAATGAAGAGAAGATCCCAAGGACAAAGAACATGAAGAGAACAA
                     ****** ***** ***** ** ** *  ** ** ******** * *****

SSIV 294/09 PA       GTCAGTTAAAGTGGGCACTTGGTGAGAATATGGCACCTGAGAAGATGGAT
SSIV 18/10 PA        GTCAGTTAGAGTGGGCACTTGGTGAAAATATGGCACCTGAGAAGGTGGAT
SSIV 247/09 PA       GTCAATTAAAGTGGGCACTTGGTGAAAATATGGCACCTGAGAAGGTGGAT
Haseluenne 2003      GTCAGCTAAAGTGGGCACTTGGTGAAAACATGGCACCTGAGAAGGTGGAT
Mexico 2009          GCCAATTGAAGTGGGCACTTGGTGAAAATATGGCACCAGAAAAAGTAGAC
                     * **  *  ***************** ** ******** ** **  * **

SSIV 294/09 PA       TTTGAAGATTGCAAAGATATCGGTGATTTAAGACAGTATGATAGTGATGA
SSIV 18/10 PA        TTTGAAGATTGCAAAGATATCGGTGATTTAAGACAGTATGATAGTGATGA
SSIV 247/09 PA       TTGAAGATTGCAAAGATATCGGTGATTTAAGACAGTATGATAGTGATGA
Haseluenne 2003      TTTGAAGATTGCAAAGATATTGGTGATTTAAAACAGTATGACAGTGATGA
Mexico 2009          TTTGATGACTGCAAAGATGTTGGAGACCTTAAACAGTATGACAGTGATGA
                     ***** ** ********* * ** **  * * ********* ********

SSIV 294/09 PA       GCCAAACAATAGATCACTGGCAAGCTGGATTCAGAGTGAATTCAATAAGG
SSIV 18/10 PA        ACCAAACAATAGATCACTGGCGAGCTGGATTCAGAGTGAATTCAACAAGG
SSIV 247/09 PA       GCCAAACAATAGATCACTGGCAAGCTGGATTCAGAGTGGATTCAACAAAG
Haseluenne 2003      GCCAAAAATTAGATCACTGGCAAGCTGGATTCAGAGTGAATTCAACAAGG
Mexico 2009          GCCAGAGCCCAGATCTCTAGCAAGCTGGGTCCAAAATGAATTCAATAAGG
                      *** *    ***** ** ** ****** * ** * ** ****** ** *

SSIV 294/09 PA       CATGTGAGTTGACGGATTCCAGCTGGATAGAGCTTGACGAAATAGGAGAG
SSIV 18/10 PA        CATGTGAGTTGACGGATTCCAGCTGGATAGAGCTTGACGAAATAGGAGAG
SSIV 247/09 PA       CATGTGAGTTGACGGATTCCAGCTGGATAGAGCTTGACGAAATAGGAGAG
Haseluenne 2003      CATGTGAGCTGACGGATTCTAGCTGGATAGAGCTTGACGAAATAGGGGAG
Mexico 2009          CATGTGAATTGACTGATTCAAGCTGGATAGAACTTGATGAAATAGGAGAA
                     *******  **** ***** ********** ***** ******** **

SSIV 294/09 PA       GATGTCGCTCCAATCGAGCACATTGCGAGTATGAGGAGGAACTACTTCAC
SSIV 18/10 PA        GATGTCGCTCCAATCGAGCACATTGCGAGTATGAGGAGGAACTACTTCAC
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SSIV 247/09 PA      GATGTCGCTCCAATCGAGCACATTGCAAGTATTAGGAGGAACTACTTCAC
Haseluenne 2003     GATGTCGCTCCAATAGAGCATATTGCGAGTATGAGGAGGAACTACTTCAC
Mexico 2009         GATGTTGCCCCGATTGAACATATCGCAAGCATGAGGAGGAACTATTTTAC
                    ***** ** ** ** ** ** ** ** ** ** *********** ** **

SSIV 294/09 PA      AGCAGAAGTGTCTCATTGCAGGGCCACGGAATACATAATGAAAGGAGTGT
SSIV 18/10 PA       AGCAGAAGTGTCTCATTGCAGGGCCACGGAATACATAATGAAAGGAGTGT
SSIV 247/09 PA      AGCAGAAGTGTCTCATTGCAGGGCCACGGAATATATAATGAAAGGAGTGT
Haseluenne 2003     AGCAGAAGTGTCTCATTGCAGGGCCACGGAGTACATAATGAAGGGAGTGT
Mexico 2009         AGCAGAAGTGTCCCACTGCAGGGCTACTGAATACATAATGAAGGGAGTGT
                    ************ ** ******** ** ** ** ******** *******

SSIV 294/09 PA      ATATAAACACAGCCTTACTCAATGCATCTTGTGCAGCAATGGATGATTTT
SSIV 18/10 PA       ATATAAACACAGCCTTACTCAATGCATCTTGTGCAGCAATGGATGATTTT
SSIV 247/09 PA      ATATAAACACAGCCTTACTCAATGCATCTTGTGCAGCAATGGATGATTTT
Haseluenne 2003     ATATAAACACAGCCTTACTCAATGCATCTTGTGCAGCAATGGATGATTTT
Mexico 2009         ACATAAATACGGCCTTGCTCAATGCATCCTGTGCAGCCATGGATGACTTT
                    * ***** ** ***** *********** ******** ******** ***

SSIV 294/09 PA      CAGTTAATCCCAATGATAAGCAAATGCAGAACGAAAGAGGGAAGGCGGAA
SSIV 18/10 PA       CAGTTAATCCCAATGATAAGCAAATGCAGAACGAAAGAAGGAAGGCGGAA
SSIV 247/09 PA      CAGTTAATCCCAATGATAAGCAAATGCAGAACGAAAGAAGGAAGGCGGAA
Haseluenne 2003     CAGTTAATCCCAATGATAAGCAAGTGCAGAACAAGAGAAGGAAGGCGGAA
Mexico 2009         CAGCTGATCCCAATGATAAGCAAATGTAGGACCAAAGAAGGAAGACGGAA
                    *** * ***************** ** ** ** * *** ***** *****

SSIV 294/09 PA      AACAAATCTATATGGATTCATTGTAAAAGGAAGATCTCATCTAAGGAACG
SSIV 18/10 PA       AACAAATCTATATGGATTCATTGTAAAAGGAAGGTCTCATCTACGGAACG
SSIV 247/09 PA      AACAAATCTATATGGGTTCATTGTAAAAGGAAGATCTCATCTAAGGAACG
Haseluenne 2003     GACAAATTTATATGGGTTCATTGTAAAAGGGAGATCTCATCTGAGAAACG
Mexico 2009         AACAAACCTGTATGGGTTCATTATAAAAGGAAGGTCTCATTTGAGAAATG
                     *****  * ***** ****** ******* ** ****** *  * ** *

SSIV 294/09 PA      ATACTGATGTGGTAAATTTTGTAAGTATGGAGTTTTCTCTCACCGATCCT
SSIV 18/10 PA       ATACTGACGTGGTAAATTTTGTAAGTATGGAGTTTTCTCTCACCGATCCT
SSIV 247/09 PA      ATACTGACGTGGTAAATTTTGTAAGTATGGAGTTTTCTCTCACCGATCCT
Haseluenne 2003     ATACCGACGTGGTAAATTTTGTGAGTATGGAGTTTTCTCTCACCGATCCG
Mexico 2009         ATACTGATGTGGTGAACTTTGTAAGTATGGAGTTCTCACTCACTGACCCG
                    **** ** ***** ** ***** *********** ** ***** ** **

SSIV 294/09 PA      AGGCTGGAGCCACACAAATGGGAGAAGTATTGTGTTCTTGAAATAGGAGA
SSIV 18/10 PA       AGGCTGGAGCCACACAAATGGGAGAAGTATTGTGTTCTTGAAATAGGAGA
SSIV 247/09 PA      AGGCTGGAGCCACACAAGTGGGAGAAGTACTGTGTTCTTGAAATAGGAGA
Haseluenne 2003     AGGCTGGAGCCACACAAGTGGGAGAAGTATTGTGTTCTTGAGATAGGAGA
Mexico 2009         AGACTGGAGCCACACAAATGGGAAAAATACTGTGTTCTTGAAATAGGAGA
                    ** ************** ***** ** ** *********** ********

SSIV 294/09 PA      CATGATTCTACGAACTTCAATTGGTCAAGTGTCAAGGCCAATGTTTCTGT
SSIV 18/10 PA       CATGATTCTACGAACTTCAATTGGCCAAGTGTCAAGGCCAATGTTTCTGT
SSIV 247/09 PA      CATGATTCTACGAACTTCAATTGGCCAAGTGTCAAGGCCAATGTTTCTAT
Haseluenne 2003     CATGATTCTACGAACTTCAATTGGCCAAGTGTCGAGGCCAATGTTTCTGT
Mexico 2009         CATGCTCTTGAGGACTGCGATAGGCCAAGTGTCGAGGCCCATGTTCCTAT
                    **** *  *  * *** * ** ** ******** ***** ***** ** *

SSIV 294/09 PA      ACGTGAGAACCAATGGAACCTCAAAAATCAAAATGAAATGGGGTATGGAA
SSIV 18/10 PA       ATGTGAGAACCAATGGAACCTCAAAAATCAAAATGAAATGGGGTGTGGAA
SSIV 247/09 PA      ATGTGAGAACCAATGGAACCTCAAAAATCAAAATGAAATGGGGTATGGAA
Haseluenne 2003     ATGTGAGAACCAATGGAACCTCAAAAATCAAAATGAAGTGGGGTATGGAA
Mexico 2009         ATGTGAGAACCAATGGAACCTCCAAGATCAAGATGAAATGGGGCATGGAA
                    * ******************** ** ***** ***** *****  *****

SSIV 294/09 PA      ATGAGGCGTTGCCTCCTTCAGTCTCTTCAACAGATTGAGAGCATGATCGA
SSIV 18/10 PA       ATGAGGCGTTGCCTCCTTCAGTCTCTTCAACAGATTGAGAGCATGATCGA
SSIV 247/09 PA      ATGAGGCGTTGCCTCCTTCAGTCTCTTCAACAGATTGAGAGCATGATCGA
Haseluenne 2003     ATGAGACGTTGCCTTCTTCAGTCCCTTCAACAGATTGAGAGCATGATCGA
Mexico 2009         CTGAGGCGCTGCCTTCTTCAGTCTCTTCAGCAGATTGAGAGCATGATTGA
                     **** ** ***** ******** ***** ***************** **

SSIV 294/09 PA      AGCGGAATCTTCCATTAAAGAAAAGGACATGACCAAAGAATTTTTTGAAA
SSIV 18/10 PA       AGCGGAATCTTCCATTAAAGAAAAGGACATGACCAAAGAATTTTTTGAAA
SSIV 247/09 PA      AGCGGAATCTTCCGTTAAAGAAAAGGACATGACCAAAGAATTTTTTGAAA
Haseluenne 2003     AGCGGAATCTTCTGTTAAAGAAAAGGACATGACCAAAGAATTTTTTGAAA
Mexico 2009         GGCCGAGTCTTCTGTCAAAGAGAAAGACATGACCAAGGAATTCTTTGAAA
                    ** ** *****  * ***** ** ********** ***** *******


SSIV 294/09 PA      ACAAATCTGAAGTATGGCCCATTGGGGAATCACCCAAAGGGGTGGAAGAA
SSIV 18/10 PA       ACAAATCTGAAGTGTGGCCCATTGGGGAATCACCCAAAGGGGTGGAAGAA
SSIV 247/09 PA      ACAAATCTGAAGTATGGCCCATTGGGGAATCACCCAAAGGGGTGGAAGAC
Haseluenne 2003     ACAAATCTGAAGCATGGCCCATTGGGGAATCACCCAAAGGGGTGGAAGAA
Mexico 2009         ACAAATCGGAAACATGGCCAATCGGAGAGTCACCCAGGGGAGTGGAGGAA
                    ******* ***   ***** ** ** ** *******  ** ***** **

SSIV 294/09 PA      GGCTCCATCGGAAAAGTATGCAGGGCCTTACTGGCAAAGTCTGTATTCAA
SSIV 18/10 PA       GGCTCCATCGGAAAAGTATGCAGGGCCTTACTGGCAAAGTCTGTATTCAA
SSIV 247/09 PA      GGCTCCATCGGAAAAGTATGCAGGGCCTTACTGGCAAAGTCTGTATTCAA
Haseluenne 2003     GGCTCCATCGGAAAAGTATGCAGGGCCTTACTGGCAAAGTCTGTATTCAA
Mexico 2009         GGCTCTATTGGGAAAGTGTGCAGGACCTTACTGGCAAAATCTGTATTCAA
                    ***** ** ** ***** ****** ************* ***********
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SSIV 294/09 PA      TAGTCTGTATGCGTCTTCACAACTCGAGGGATTTTCAGCCGAATCAAGAA
SSIV 18/10 PA       TAGTCTGTATGCGTCTTCACAACTCGAGGGATTTTCAGCTGAATCAAGAA
SSIV 247/09 PA      TAATCTGTATGCGTCTTCACAACTCGAGGGATTTTCAGCTGAATCAAGAA
Haseluenne 2003     TAGCCTGTATGCATCTCCACAACTCGAGGGATTCTCAGCTGAATCAAGAA
Mexico 2009         CAGTCTATATGCGTCTCCACAACTTGAGGGGTTTTCGGCTGAATCGAGAA
                     *  ** ***** *** ******* ***** ** ** ** ***** ****

SSIV 294/09 PA      AGTTACTTCTAATTGTTCAGGCACTTAGGGACAACCTGGAACCTGGGACC
SSIV 18/10 PA       AGTTACTTCTAATTGTTCAGGCACTTAGGGACAACCTGGAACCTGGGACC
SSIV 247/09 PA      AGTTACTTCTAATTGTTCAGGCACTTAGGGACAACCTGGAACCTGGGACC
Haseluenne 2003     AGTTGCTTCTAATTGTTCAGGCACTTAGGGACAACCTGGAACCTGGGACC
Mexico 2009         AATTGCTTCTCATTGTTCAGGCACTTAGGGACAACCTGGAACCTGGAACC
                    * ** ***** ********************************** ***

SSIV 294/09 PA      TTTGATCTTGGGGGATTATATGATGCAATTGAGGAGTGCCTGATTAATGA
SSIV 18/10 PA       TTTGATCTTGGGGGATTATATGATGCAATTGAGGAGTGCCTGATTAATGA
SSIV 247/09 PA      TTTGATCTTGGGGGATTATATGATGCAATTGAGGAGTGCCTGATTAATGA
Haseluenne 2003     TTTGATCTTGAAGGACTATATGATGCAATTGAGGAGTGCCTGATTAATGA
Mexico 2009         TTCGATCTTGGGGGGCTATATGAAGCAATCGAGGAGTGCCTGATTAATGA
                    ** *******  **  ******* ***** ********************

SSIV 294/09 PA      TCCCTGGGTTTTGCTTAATGCGTCTTGGTTCAACTCCTTCCTTATTCATG
SSIV 18/10 PA       TCCCTGGGTTTTGCTTAATGCGTCTTGGTTCAACTCCTTCCTTATTCATG
SSIV 247/09 PA      TCCCTGGGTTTTGCTTAACGCGTCTTGGTTCAACTCCTTCCTTATTCATG
Haseluenne 2003     TCCCTGGGTTTTGCTTAATGCGTCTTGGTTCAACTCCTTCCTTGTGCATG
Mexico 2009         TCCCTGGGTTTTGCTTAATGCATCTTGGTTCAACTCCTTCCTCACACATG
                    ****************** ** ********************    ****

SSIV 294/09 PA      CACTGAAATAATTGTGGCAATGCTACTACTTGCTATCCATACTGTCCAAA
SSIV 18/10 PA       CACTGAAATAATTGTGGCAATGCTACTACTTGCTATCCATACTGTCCAAA
SSIV 247/09 PA      CACTGAAATAATTGTGGCAATGCTACTACTTGCTATCCATACTGTCCAAA
Haseluenne 2003     CACTGAAATAGTTGTGGCAATGCTACTACTTGCTATCCATACTGTCCAAA
Mexico 2009         CACTGAAGTAGTTGTGGCAATGCTACTAT----------------------
                    ******* ** ******************

SSIV 294/09 PA      AAAGTACCTTGTTTCNANTAATACGAGAC
SSIV 18/10 PA       AAAGTACCTTGTTTCTACTAATACGAGAC
SSIV 247/09 PA      AAAGTACCTTGTTTCTACTAATACGAGAC
Haseluenne 2003     AAAGTACCTTGTTTCTACT----------
Mexico 2009         ----------------------------
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Legend:
Nucleotide sequences of segment 3. Three positions causing significant differences on the aa level are highlighted red.

Appendix 6b

PA protein

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SSIV 18/10 PA       MEDFVRQCFNPMIVELAEKIMKEHGEDPKIETNKFAAICTHMEVCFMYSD
SSIV 247/09 PA      MEDFVRQCFNPMIVELAEKIMKEHGEDPKIEINKFAAICTHMEVCFMYSD
SSIV 294/09 PA      MEDFVRQCFNPMIVELAEKIMKEHGEDPKIETNKFAAICTHMEVCFMYSD
Haseluenne 2003     MEDFVRQCFNPMIVELAEKAMKEHGEDPKIETNKFAAICTHLEVCFMYSD
Mexico 2009         MEDFVRQCFNPMIVELAEKAMKEYGEDPKIETNKFAAICTHLEVCFMYSD
                    ****************** ***:******* *********:********

SSIV 18/10 PA       FHSIDERGESTIVESSDPNALLKHRFEIIEGRDRTVAWTVVNSICNTTGV
SSIV 247/09 PA      FHFIDERGESTIVESSDPNALLKHRFEIIEGRDRTVAWTVVNSICNTTGV
SSIV 294/09 PA      FHFIDERGESTIVESSDPNALLKHRFEIIEGRDRTVAWTVVNSICNTTGV
Haseluenne 2003     FHFIDERGESIVVESGDPNALLKHRFEIIEGRDRTMAWTVVNSICNTTEV
Mexico 2009         FHFIDERGESIIVESGDPNALLKHRFEIIEGRDRIMAWTVVNSICNTTGV
                    ** ******* :***.****************** :************ *

SSIV 18/10 PA       EKPKFLPDLYDYKENRFIEIGVTRRGVHIYYLEKANKIKSEKTHIHIFSF
SSIV 247/09 PA      EKPKFLPDLYDYKENRFIEIGVTRREVHIYYLEKANKIKSEKTHIHIFSF
SSIV 294/09 PA      EKPKFLPDLYDYKENRFIEIGVTRREVHIYYLEKANKIKSEKTHIHIFSF
Haseluenne 2003     EKPKFLPDLYDYKENRFIEIGVTRREVHIYYLEKANKIKSEKTHIHIFSF
Mexico 2009         EKPKFLPDLYDYKENRFIEIGVTRREVHIYYLEKANKIKSEKTHIHIFSF
                    ************************ ************************

SSIV 18/10 PA       TGEEMATKADYTLDEESRARIKTRLFTIRQEMAIRALWDSFRQSERGEET
SSIV 247/09 PA      TGEEMATKADYTLDEESRARIKTRLFTIRQEMAIRALWDSFRQSERGEET
SSIV 294/09 PA      TGEEMATKADYTLDEESRARIKTRLFTIRQEMAIRALWDSFRQSERGEET
Haseluenne 2003     TGEEMATKADYTLDEESRARIKTRLFTIRQELAIRGLWDSFRQSERGEET
Mexico 2009         TGEEMATKADYTLDEESRARIKTRLFTIRQEMASRSLWDSFRQSERGEET
                    ******************************:* *.***************

SSIV 18/10 PA       IEERFEIKGTMHRLANQSLPPNFSSIENFRAYVDGFEPNGCIEGKLSQMS
SSIV 247/09 PA      IEERFEIKGTMHRLANQSLPPNFSSLENFRAYVDGFEPNGCIEGKLSQMS
SSIV 294/09 PA      IEERFEIKGTMHRLANQSLPPNFSSLENFRAYVDGFEPNGCIEGKLSQMS
Haseluenne 2003     IEERFEIKGTMRRLANQSLPPNFSSLENFRAYVDGFEPNSCIESKLSQMS
Mexico 2009         IEEKFEITGTMRKLADQSLPPNFSSLENFRAYVDGFEPNGCIEGKLSQMS
                    ***:***.***::**:*********:*************.***.******
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SSIV 18/10 PA      KEVNARIEPFLRGTPRPLRLPDGPPCSQRSKFLLMDALKLSIEDPSHEGE
SSIV 247/09 PA     KEVNARIEPFLRGTPRPLRLPDGPPCSQRSKFLLMDALKLSIEDPSHEGE
SSIV 294/09 PA     KEVNARIEPFLRGTPRPLRLPDGPLCSQRSKFLLMDALKLSIEDPSHEGE
Haseluenne 2003    KEVNARIDPFLRETPRPLRLPDGPPCSQRSKFLLMDALKLSIEDPSHEGE
Mexico 2009        KEVNAKIEPFLRTTPRPLRLPDGPLCHQRSKFLLMDALKLSIEDPSHEGE
                   *****:*:**** ********** * **********************

SSIV 18/10 PA      GIPLYDAIKCMKTFFGWKEPHIVKPHEKGINPNYLLAWKQILAELQDIEN
SSIV 247/09 PA     GIPLYDALKCMKTFFGWKEPHIVKPHEKGINPNYLLAWKQILAELQDIEN
SSIV 294/09 PA     GIPLYDAIKCMKTFFGWKEPHIVKPHERGINPNYLLAWKQILAELQDIEN
Haseluenne 2003    GIPLYDAIKCMKTFFGWKEPNIVKPHEKGINPNYLLAWKQVLAEIQDIEN
Mexico 2009        GIPLYDAIKCMKTFFGWKEPNIVKPHEKGINPNYLMAWKQVLAELQDIEN
                   *******:************:******:*******:****:***:*****

SSIV 18/10 PA      EEKIPKTKNMKKTSQLEWALGENMAPEKVDFEDCKDIGDLRQYDSDEPNN
SSIV 247/09 PA     EEKIPKTKNMKKTSQLKWALGENMAPEKVDFEDCKDIGDLRQYDSDEPNN
SSIV 294/09 PA     EEKIPKTKNMKKTSQLKWALGENMAPEKMDFEDCKDIGDLRQYDSDEPNN
Haseluenne 2003    EEKIPKTKNMKKTSQLKWALGENMAPEKVDFEDCKDIGDLKQYDSDEPKI
Mexico 2009        EEKIPRTKNMKRTSQLKWALGENMAPEKVDFDDCKDVGDLKQYDSDEPEP
                   *****:*****:****:***********:**:****:***:*******:

SSIV 18/10 PA      RSLASWIQSEFNKACELTDSSWIELDEIGEDVAPIEHIASMRRNYFTAEV
SSIV 247/09 PA     RSLASWIQSGFNKACELTDSSWIELDEIGEDVAPIEHIASIRRNYFTAEV
SSIV 294/09 PA     RSLASWIQSEFNKACELTDSSWIELDEIGEDVAPIEHIASMRRNYFTAEV
Haseluenne 2003    RSLASWIQSEFNKACELTDSSWIELDEIGEDVAPIEHIASMRRNYFTAEV
Mexico 2009        RSLASWVQNEFNKACELTDSSWIELDEIGEDVAPIEHIASMRRNYFTAEV
                   ******:*. ****************************:*********

SSIV 18/10 PA      SHCRATEYIMKGVYINTALLNASCAAMDDFQLIPMISKCRTKEGRRKTNL
SSIV 247/09 PA     SHCRATEYIMKGVYINTALLNASCAAMDDFQLIPMISKCRTKEGRRKTNL
SSIV 294/09 PA     SHCRATEYIMKGVYINTALLNASCAAMDDFQLIPMISKCRTKEGRRKTNL
Haseluenne 2003    SHCRATEYIMKGVYINTALLNASCAAMDDFQLIPMISKCRTREGRRKTNL
Mexico 2009        SHCRATEYIMKGVYINTALLNASCAAMDDFQLIPMISKCRTKEGRRKTNL
                   ****************************************:********

SSIV 18/10 PA      YGFIVKGRSHLRNDTDVVNFVSMEFSLTDPRLEPHKWEKYCVLEIGDMIL
SSIV 247/09 PA     YGFIVKGRSHLRNDTDVVNFVSMEFSLTDPRLEPHKWEKYCVLEIGDMIL
SSIV 294/09 PA     YGFIVKGRSHLRNDTDVVNFVSMEFSLTDPRLEPHKWEKYCVLEIGDMIL
Haseluenne 2003    YGFIVKGRSHLRNDTDVVNFVSMEFSLTDPRLEPHKWEKYCVLEIGDMIL
Mexico 2009        YGFIIKGRSHLRNDTDVVNFVSMEFSLTDPRLEPHKWEKYCVLEIGDMLL
                   ****:*******************************************:*

SSIV 18/10 PA      RTSIGQVSRPMFLYVRTNGTSKIKMKWGVEMRRCLLQSLQQIESMIEAES
SSIV 247/09 PA     RTSIGQVSRPMFLYVRTNGTSKIKMKWGMEMRRCLLQSLQQIESMIEAES
SSIV 294/09 PA     RTSIGQVSRPMFLYVRTNGTSKIKMKWGMEMRRCLLQSLQQIESMIEAES
Haseluenne 2003    RTSIGQVSRPMFLYVRTNGTSKIKMKWGMEMRRCLLQSLQQIESMIEAES
Mexico 2009        RTAIGQVSRPMFLYVRTNGTSKIKMKWGMELRRCLLQSLQQIESMIEAES
                   **:*************************:*:******************


SSIV 18/10 PA      SIKEKDMTKEFFENKSEVWPIGESPKGVEEGSIGKVCRALLAKSVFNSLY
SSIV 247/09 PA     SVKEKDMTKEFFENKSEVWPIGESPKGVEDGSIGKVCRALLAKSVFNNLY
SSIV 294/09 PA     SIKEKDMTKEFFENKSEVWPIGESPKGVEEGSIGKVCRALLAKSVFNSLY
Haseluenne 2003    SVKEKDMTKEFFENKSEAWPIGESPKGVEEGSIGKVCRALLAKSVFNSLY
Mexico 2009        SVKEKDMTKEFFENKSETWPIGESPRGVEEGSIGKVCRTLLAKSVFNSLY
                   *:***************.*******:***:********:********.**

SSIV 18/10 PA     ASSQLEGFSAESRKLLLIVQALRDNLEPGTFDLGGLYDAIEECLINDPWV
SSIV 247/09 PA    ASSQLEGFSAESRKLLLIVQALRDNLEPGTFDLGGLYDAIEECLINDPWV
SSIV 294/09 PA    ASSQLEGFSAESRKLLLIVQALRDNLEPGTFDLGGLYDAIEECLINDPWV
Haseluenne 2003   ASPQLEGFSAESRKLLLIVQALRDNLEPGTFDLEGLYDAIEECLINDPWV
Mexico 2009       ASPQLEGFSAESRKLLLIVQALRDNLEPGTFDLGGLYEAIEECLINDPWV
                  **.***************************** ***:************

SSIV 18/10 PA     LLNASWFNSFLIHALK
SSIV 247/09 PA    LLNASWFNSFLIHALK
SSIV 294/09 PA    LLNASWFNSFLIHALK
Haseluenne 2003   LLNASWFNSFLVHALK
Mexico 2009       LLNASWFNSFLT----
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Legend:
Amino acid sequences of the PA protein. Position 20 highlighted in different colors. There, alanine is considered typical for influenza A viruses, thr typical for influenza B viruses, and ile typical for influenza C viruses.

Appendix 7a

Segment 2

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SSIV 18/10 PB1     ATAGGGCGAATTGGGCCCTCTAGATGCATGCTCGAGCGGCCGCCAGTGTG
SSIV 294/09 PB1    ATAGGGCGAATTGGGCCCTCTAGATGCATGCTCGAGCGGCCGCCAGTGTG
SSIV 247/09 PB1    ATAGGGCGAATTGGGCCCTCTAGATGCATGCTCGAGCGGCCGCCAGTGTG
Haseluenne 2003    ------------------------------------------------
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Mexico 2009       --------------------------------------------------


SSIV 18/10 PB1    ATGGATATCTGCAGAATTCGCCCTTTATTCGTCTCAGGGAGCGAAAGCAG
SSIV 294/09 PB1   ATGGATATCTGCAGAATTCGCCCTTTATTCGTCTCAGGGAGCGAAAGCAG
SSIV 247/09 PB1   ATGGATATCTGCAGAATTCGCCCTTTATTCGTCTCAGGGAGCGAAAGCAG
Haseluenne 2003   ---------------------------------------AGCGAAAGCAG
Mexico 2009       --------------------------------------------------


SSIV 18/10 PB1    GCAAACCATTTGAATGGATGTCAACCCGACTCTACTTTTCTTAAAAGTGC
SSIV 294/09 PB1   GCAAACCATTTGAATGGATGTCAACCCGACTCTACTTTTCTTAAAAGTGC
SSIV 247/09 PB1   GCAAACCATTTGAATGGATGTCAACCCGATTCTACTTTTCTTAAAAGTGC
Haseluenne 2003   GCAAACCATTTGAATGGATGTCAACCCGACTCTACTTTTCTTAAAAGTGC
Mexico 2009       ---------TTGAATGGATGTCAATCCGACTCTACTTTTCCTAAAAATTC
                            *************** **** ********** ***** * *


SSIV 18/10 PB1    CAGCACAAAATGCTATAAGCACTACATTCCCTTATACTGGAGATCCCCCA
SSIV 294/09 PB1   CAGCACAAAATGCTATAAGCACTACATTCCCTTATACTGGAGATCCCCCA
SSIV 247/09 PB1   CAGCACAAAATGCTATAAGCACTACATTCCCTTACACTGGAGATCCCCCA
Haseluenne 2003   CAGCACAAAATGCTATAAGCACTACATTCCCTTATACTGGAGATCCTCCA
Mexico 2009       CAGCGCAAAATGCCATAAGCACCACATTCCCTTATACTGGAGATCCTCCA
                  **** ******** ******** *********** *********** ***


SSIV 18/10 PB1    TACAGCCATGGAACAGGGACTGGATACACCATGGACACTGTCAACAGAAC
SSIV 294/09 PB1   TACAGTCATGGAACAGGGACTGGATACACCATGGACACTGTCAACAGAAC
SSIV 247/09 PB1   TACAGTCATGGAACAGGGACTGGATACACCATGGACACGGTCAACAGAAC
Haseluenne 2003   TATAGTCATGGAACAGGGACAGGATACACCATGGACACGGTCAACAGAAC
Mexico 2009       TACAGCCATGGAACAGGAACAGGATACACCATGGACACAGTAAACAGAAC
                  ** ** *********** ** ***************** ** ********


SSIV 18/10 PB1    ACATCAATATTCAGAAAGGGGAAGATGGACAACAAACACAGAAACCGGGG
SSIV 294/09 PB1   ACATCAATATTCAGAAAGGGGAAGATGGACAACAAACACAGAAACTGGAG
SSIV 247/09 PB1   ACATCAATATTCAGAAAGGGGAAAATGGACAACAAACACAGAAACTGGAG
Haseluenne 2003   ACATCAATATTCAGAAAGGGGAAGGTGGACAACAAACACAGAAACTGGAG
Mexico 2009       ACACCAATACTCAGAAAAGGGAAAGTGGACGACAAACACAGAGACTGGTG
                  *** ***** ******* *****  ***** *********** ** ** *


SSIV 18/10 PB1    CACCCCAGCTTAACCCGATTGACGGACCACTACCTGAGGACAATGAGCCA
SSIV 294/09 PB1   CACCCCAACTTAACCCGATTGACGGACCACTACCTGAGGACAATGAGCCA
SSIV 247/09 PB1   CACCCCAGCTTAACCCGATTGATGGACCACTACCTGAGGACAATGAGCCA
Haseluenne 2003   CACCCCAGCTTAACCCGATTGATGGACCACTACCTGAGGACAATGAGCCA
Mexico 2009       CACCCCAGCTCAACCCGATTGATGGACCACTACCTGAGGATAATGAACCA
                  ******* ** *********** ***************** ***** ***


SSIV 18/10 PB1    AGCGGATATGCTCAAACAGACTGTGTCCTGGAGGCAATGGCTTTCCTTGA
SSIV 294/09 PB1   AGCGGATATGCTCAAACAGACTGTGTCCTGGAGGCGATGGCTTTCCTTGA
SSIV 247/09 PB1   AGCGGGTATGCTCAAACAGACTGTGTCCTGGAGGCAATGGCTTTCCTTGA
Haseluenne 2003   AGCGGGTATGCTCAAACAGACTGTGTCCTGGAGGCAATGGCTTACCTTGA
Mexico 2009       AGTGGGTATGCACAAACAGACTGTGTTCTAGAGGCTATGGCTTTCCTTGA
                  ** ** ***** ************** ** ***** ******* ******


SSIV 18/10 PB1    GGAATCCCATCCAGGGATATTTGAAAACTCTTGTCTTGAAACGATGGAGG
SSIV 294/09 PB1   GGAATCCCATCCAGGGATATTTGAAAACTCTTGTCTTGAAACGATGGAGG
SSIV 247/09 PB1   GGAATCCCATCCAGGGATATTTGAAAACTCTTGTCTTGAAACGATAGAGG
Haseluenne 2003   GGAATCCCACCCAGGGATATTCGAAAACTCGTGTCTTGAAACAATGGAAG
Mexico 2009       AGAATCCCACCCAGGAATATTTGAGAATTCATGCCTTGAAACAATGGAAG
                  ******** ***** ***** ** ** ** ** ******** ** ** *


SSIV 18/10 PB1    TTGTCCAACAAACAAGAGTGGACAAACTGACTCAAGGTCGTCAGACCTAT
SSIV 294/09 PB1   TTGTCCAACAAACAAGAGTGGACAAACTGACTCAAGGTCGTCAGACCTAT
SSIV 247/09 PB1   TTGTCCAACAAACAAGAGTGGACAAACTGACTCAAGGTCGTCAGACCTAT
Haseluenne 2003   TTGTCCAACAAACAAGAGTGGACAAACTGACTCAAGGTCGTCAGACCTAT
Mexico 2009       TTGTTCAACAAACAAGGGTAGATAAACTAACTCAAGGTCGCCAGACTTAT
                  **** *********** ** ** ***** *********** ***** ***


SSIV 18/10 PB1    GGCTGGACATTGAATAGAAACCAACCTGCTGCAACTGCTTTGGCCAACAC
SSIV 294/09 PB1   GACTGGACATTGAATAGAAACCAACCTGCTGCAACTGCTTTGGCCAACAC
SSIV 247/09 PB1   GACTGGACATTGAATAGAAACCAACCTGCTGCAACTGCTTTGGCCAACAC
Haseluenne 2003   GACTGGACATTGAATAGAAACCAACCAGCTGCAACTGCTTTGGCCAACAC
Mexico 2009       GATTGGACATTAAACAGAAATCAACCGGCAGCAACTGCATTGGCCAACAC
                  *  ******** ** ***** ***** ** ******** ***********


SSIV 18/10 PB1    AATAGAAGTCTTCAGGATGAACAGTCTAACAGCCAATGAATCGAGAAGAT
SSIV 294/09 PB1   AATAGAAGTCTTCAGGATGAACAGTCTAACAGCCAATGAATCGGGAAGAT
SSIV 247/09 PB1   AATAGAAGTCTTCAGGATGAACAGTCTAACAGCCAATGAATCGGGAAGAT
Haseluenne 2003   AATAGAGGTCTTCAGGTTGAACAGCCTAACAGCCAATGAATCGGGGAGAT
Mexico 2009       CATAGAAGTCTTTAGATCGAATGGCCTAACAGCTAATGAGTCAGGAAGGC
                  ***** ***** **   ***  * ******** ***** **  * **


SSIV 18/10 PB1    TAATAGATTTCCTTAAGGACGTGATGGAATCAATGGACAAAGAAGAGATG
SSIV 294/09 PB1   TAATAGATTTCCTTAAGGACGTGATGGAATCAATGGACAAAGAAGAGATG
SSIV 247/09 PB1   TAATAGATTTCCTTAAGGACGTGATGGAATCAATGGACAAAGAAGAGATG
Haseluenne 2003   TAATAGATTTCCTTAAAGATGTGATGGAATCAATGGATAAAGAAGAGATG
Mexico 2009       TAATAGATTTCTTAAAGGATGTAATGGAATCAATGAACAAAGAGGAAATA
                  *********** * ** ** ** ************ * ***** ** **


SSIV 18/10 PB1    GAAATAACAACACATTTCCAAAGGAAAAGAAGAATAAGGGACAACATGAC
SSIV 294/09 PB1   GAAATAACAACACATTTCCAAAGGAAAAGAAGAATAAGGGACAACATGAC
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SSIV 247/09 PB1   GAAATAACAACACATTTCCAAAGGAAAAGAAGAATAAGGGACAACATGAC
Haseluenne 2003   GAAATAACAACACATTTCCAAAGGAAAAGAAGAATAAGGGACAACATGAC
Mexico 2009       GAGATAACAACCCACTTTCAAAGAAAAAGGAGAGTAAGAGACAACATGAC
                  ** ******** ** ** ***** ***** *** **** ***********

SSIV 18/10 PB1    CAAGAAAATGGTAACACAAAGGACAATAGGAAAGAAGAAGCAGAAGCTGA
SSIV 294/09 PB1   CAAGAAAATGGTAACACAAAGAACAATAGGAAAGAAGAAGCAGAAGCTGA
SSIV 247/09 PB1   AAAGAAAATGGTAACACAAAGGACAATAGGAAAGAAGAAGCAGAAACTGA
Haseluenne 2003   CAAGAAAATGATAACACAAAGGACAATAGGGAAGAAAAAGCAGAAACTGA
Mexico 2009       CAAGAAGATGGTCACGCAAAGAACAATAGGGAAGAAAAAACAAAGACTGA
                  ***** *** * ** ***** ******** ***** ** ** *  ****

SSIV 18/10 PB1    ACAAAAAGAATTATCTAATAAGAGCATTGACATTAAACACAATGACAAAA
SSIV 294/09 PB1   ACAAGAAGAATTATCTAATAAGAGCATTGACATTAAACACAATGACAAAA
SSIV 247/09 PB1   ACAAGAAGAATTATCTAATAAGAGCATTGACATTAAACACAATGACAAAA
Haseluenne 2003   ACAAGAGGAGTTATCTAATAAGAGCATTGACATTAAACACAATGACAAAA
Mexico 2009       ATAAGAGAGGCTATCTAATAAGAGCACTGACATTAAATACGATGACCAAA
                  * ** *     *************** ********** ** ***** ***

SSIV 18/10 PB1   GATGCAGAAAGAGGCAAGTTGAAAAGGCGTGCAATTGCAACACCAGGGAT
SSIV 294/09 PB1  GATGCAGAAAGAGGCAAGTTGAAAAGGCGTGCAATTGCAACACCAGGGGT
SSIV 247/09 PB1  GATGCAGAAAGAGGCAAGTTGAAAAGGCGTGCAATTGCAACACCAGGGAT
Haseluenne 2003  GATGCAGAAAGAGGCAAGTTGAAAAGGCGTGCAATTGCAACACCAGGGAT
Mexico 2009      GATGCAGAGAGAGGCAAGTTAAAAAGAAGGGCTATCGCAACACCTGGGAT
                 ******** *********** *****  * ** ** ******** *** *

SSIV 18/10 PB1    GCAGATCAGAGGATTTGTGTACTTCGTAGAAACACTGGCGAGGAGCATCT
SSIV 294/09 PB1   GCAGATCAGAGGATTTGTGTACTTCGTAGAAACACTGGCGAGGAGCATCT
SSIV 247/09 PB1   GCAGATCAGAGGATTTGTGTACTTCGTAGAAACACTGGCGAGGAGCATCT
Haseluenne 2003   GCAGATCAGAGGGTTTGTGTACTTTGTAGAAACACTGGCTAGGAGCATCT
Mexico 2009       GCAGATTAGAGGTTTCGTATACTTTGTTGAAACTTTAGCTAGGAGCATTT
                  ****** ***** ** ** ***** ** *****  * ** ******** *

SSIV 18/10 PB1    GTGAGAAACTTGAGCAATCCGGGCTCCCAGTTGGAGGGAATGAGAAGAAA
SSIV 294/09 PB1   GTGAGAAACTTGAGCAATCCGGGCTCCCAGTTGGAGGGAATGAGAAGAAA
SSIV 247/09 PB1   GTGAGAAACTTGAGCAATCCGGGCTCCCAGTTGGGGGGAATGAGAAGAAA
Haseluenne 2003   GTGAGAAACTTGAGCAATCTGGGCTCCCAGTTGGAGGGAATGAAAAGAAA
Mexico 2009       GCGAAAAGCTTGAACAGTCTGGGCTCCCAGTAGGGGGCAATGAAAAGAAG
                  * ** ** ***** ** ** *********** ** ** ***** *****

SSIV 18/10 PB1    GCCAAATTGGCAAATGTTGCGAGGAAAATGATGACCAACTCACAAGATAC
SSIV 294/09 PB1   GCCAAATTGGCAAATGTTGTGAGGAAAATGATGACCAACTCACAAGATAC
SSIV 247/09 PB1   GCCAAATTGGCAAATGTTGTGAGAAAAATGATGACCAACTCACAAGATAC
Haseluenne 2003   GCCAAATTGGCAAATGTTGTGAGGAAAATGATGACAAACTCACAAGATAC
Mexico 2009       GCCAAACTGGCAAATGTTGTGAGAAAGATGATGACTAATTCACAAGACAC
                  ****** ************ *** ** ******** ** ******** **

SSIV 18/10 PB1    AGAGCTTTCCTTTACAATTACTGGGGACAACACCAAATGGAATGAGAATC
SSIV 294/09 PB1   AGAGCTTTCCTTTACAATTACTGGGGACAACACCAAATGGAATGAGAATC
SSIV 247/09 PB1   AGAGCTTTCCTTTACAATTACTGGGGACAACACCAAATGGAATGAGAATC
Haseluenne 2003   AGAGCTCTCCTTTACAATTACTGGAGACAACACCAAATGGAATGAGAATC
Mexico 2009       AGAGATTTCTTTCACAATCACTGGGGACAACACTAAGTGGAATGAAAATC
                  **** * ** ** ***** ***** ******** ** ******** ****

SSIV 18/10 PB1    AAAACCCCAGGATGTTTCTGGCGATGATAACGTATGTCACAAGAAATCAG
SSIV 294/09 PB1   AAAACCCCAGGATGTTTCTGGCGATGATAACGTATATCACAAGAAATCAG
SSIV 247/09 PB1   AAAACCCTAGGATGTTTCTGGCGATGATAACGTATATCACAAGAAATCAG
Haseluenne 2003   AAAACCCACGGGTGTTTCTGGCTATGATAACATACATCACAAGAAATCAG
Mexico 2009       AAAATCCTCGAATGTTCCTGGCGATGATTACATATATCACCAGAAATCAA
                  **** **  *  **** ***** ***** ** **  **** ********

SSIV 18/10 PB1    CCTGAATGGTTCAGAAATGTTTTAAGCATCGCCCCTATAATGTTCTCAAA
SSIV 294/09 PB1   CCTGAATGGTTCAGAAATGTTTTAAGCATCGCCCCTATAATGTTCTCAAA
SSIV 247/09 PB1   CCTGAATGGTTCAGAAATGTTTTAAGCATCGCCCCTATAATGTTCTCAAA
Haseluenne 2003   CCTGAATGGTTCAGAAATGTTTTAAGCATCGCCCCTATAATGTTCTCAAA
Mexico 2009       CCCGAGTGGTTCAGAAACATCCTGAGCATGGCACCCATAATGTTCTCAAA
                  ** ** ***********  *  * ***** ** ** **************

SSIV 18/10 PB1    CAAAATGGCAAGATTAGGAAAAGGATACATGTTCGAAAGCAAGAGCATGA
SSIV 294/09 PB1   CAAAATGGCAAGATTAGGAAAAGGATACATGTTCGAAAGCAAGAGCATGA
SSIV 247/09 PB1   CAAAATGGCAAGATTAGGGAAAGGATACATGTTCGAGAGTAAGAGCATGA
Haseluenne 2003   TAAAATGGCAAGATTAGGGAAAGGATACATGTTCGAAAGTAAGAGCATGA
Mexico 2009       CAAAATGGCAAGACTAGGGAAAGGGTACATGTTCGAGAGTAAAAGAATGA
                  ************ **** ***** *********** ** ** ** ****

SSIV 18/10 PB1    AGCTACGAGCACAAATACCAGCAGAAATGCTCGCTGATATTGACTTGAAA
SSIV 294/09 PB1   AGCTACGAACACAGATACCAGCAGAAATGCTCGCAGATATTGACTTGAAA
SSIV 247/09 PB1   AGCTACGAACACAAATACCAGCAGAAATGCTCGCAGATATTGACTTGAAA
Haseluenne 2003   AGCTACGAACACAAATACCAGCAGAAATGCTAGCAGATATTGACTTGAAA
Mexico 2009       AGATTCGAACACAAATACCAGCAGAAATGCTAGCAAGCATTGACCTGAAG
                  ** * *** **** ***************** **    ****** ****

SSIV 18/10 PB1    TACTTCAATGGGTCAACAAGGAAAAAGATCGAGAAGATAAGGCCACTCCT
SSIV 294/09 PB1   TACTTCAATGAGTCAACAAGGAAAAAGATCGAGAAGATAAGGCCACTCCT
SSIV 247/09 PB1   TACTTCAATGAGTCAACAAGGAAAAAGATCGAGAAGATAAGGCCACTCCT
Haseluenne 2003   TACTTCAATGAATCAACAAGGAAAAAGATCGAGAAGATAAGGCCACTTCT
Mexico 2009       TACTTCAATGAATCAACAAAGAAGAAAATTGAGAAAATAAGGCCTCTTCT
                  **********  ******* *** ** ** ***** ******** ** **
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SSIV 18/10 PB1    CATAGATGGAACGGCCTCTCTAAGCCCTGGAATGATGATGGGCATGTTTA
SSIV 294/09 PB1   CATAGATGGAACGGCCTCACTAAGCCCTGGAATGATGATGGGCATGTTTA
SSIV 247/09 PB1   CATAGATGGCACGGCCTCACTGAGCCCTGGAATGATGATGGGCATGTTTA
Haseluenne 2003   AATAAATGGCACGGCCTCATTAAGCCCTGGAATGATGATGGGCATGTTTA
Mexico 2009       AATAGATGGCACAGCATCACTGAGTCCTGGGATGATGATGGGCATGTTCA
                  *** **** ** ** **  * ** ***** ***************** *

SSIV 18/10 PB1    ACATGCTGAGCACAGTTCTAGGAGTTTCAATCCTAAACCTCGGGCAAAAG
SSIV 294/09 PB1   ACATGCTGAGCACAGTTCTAGGAGTTTCAATCCTAAACCTCGGGCAAAAG
SSIV 247/09 PB1   ACATGCTGAGCACAGTTCTGGGAGTTTCAATCCTGAACCTCGGGCAAAAG
Haseluenne 2003   ACATGCTGAGCACAGTCCTAGGAGTCTCGATCCTGAACCTTGGGCAAAAG
Mexico 2009       ACATGCTAAGTACGGTCTTGGGAGTCTCAATACTGAATCTTGGACAAAAG
                  ** ** **  * ***** ** ** ** ** ** ** ******

SSIV 18/10 PB1    AGGTACACTAAAACCACCTACTGGTGGGATGGACTCCAATCTTCTGATGA
SSIV 294/09 PB1   AGGTACACTAAAACCACCTACTGGTGGGATGGACTCCAATCTTCTGATGA
SSIV 247/09 PB1   AGGTACACTAAAACCACCTACTGGTGGGATGGACTCCAATCTTCTGATGA
Haseluenne 2003   AGGTACACTAAAACCACATACTGGTGGGATGGACTCCAATCTTCTGATGA
Mexico 2009       AAATACACCAAGACAATATACTGGTGGGATGGGCTCCAATCATCCGACGA
                  *  ***** ** ** *  ************** ******** ** ** **

SSIV 18/10 PB1    CTTCGCCCTCATAGTGAATGCACCTAATCACGAAGGAATACAAGCAGGAG
SSIV 294/09 PB1   CTTCGCCCTCATAGTGAATGCACCTAATCACGAAGGAATACAAGCAGGAG
SSIV 247/09 PB1   CTTCGCCCTCATAGTGAATGCACCTAATCACGAAGGAATACAGGCAGGAG
Haseluenne 2003   CTTCGCCCTCATAGTGAATGCACCTAGTCATGAAGGAATACAAGCAGGAG
Mexico 2009       TTTTGCTCTCATAGTGAATGCACCAAACCATGAGGGAATACAAGCAGGAG
                   ** ** ****************** *  ** ** ******** *******

SSIV 18/10 PB1    TGGATAGGTTCTATAGGACCTGCAAACTAGTTGGAATCAATATGAGCAAA
SSIV 294/09 PB1   TGGATAGGTTCTATAGGACCTGCAAATTAGTTGGAATCAATATGAGCAAA
SSIV 247/09 PB1   TGGATAGGTTCTACAGGACCTGCAAACTGGTTGGAATCAACATGAGCAAA
Haseluenne 2003   TAGATAGGTTCTATAGGACCTGCAAACTGGTTGGAATCAACATGAGCAAA
Mexico 2009       TGGACAGATTCTACAGGACCTGCAAGTTAGTGGGAATCAACATGAGCAAA
                  * ** ** ***** ***********  * ** ******** *********

SSIV 18/10 PB1    AAGAAATCTTATATAAATAGGACAGGAACATTTGAATTCACAAGCTTTTT
SSIV 294/09 PB1   AAGAAATCTTATATAAATAGGACAGGAACATTTGAATTCACAAGCTTTTT
SSIV 247/09 PB1   AAGAAATCTTATATAAATAGGACAGGAACATTTGAATTCACAAGCTTTTT
Haseluenne 2003   AAGAAATCTTACATAAATAGGACAGGAACATTTGAATTCACAAGCTTTTT
Mexico 2009       AAGAAGTCCTATATAAATAAGACAGGGACATTTGAATTCACAAGCTTTTT
                  ***** ** ** ******* ****** ***********************

SSIV 18/10 PB1    CTACCGCTATGGGTTTGTAGCCAACTTCAGTATGGAGCTACCTAGCTTTG
SSIV 294/09 PB1   CTACCGCTATGGGTTTGTAGCCAACTTCAGTATGGAGCTACCTAGCTTTG
SSIV 247/09 PB1   CTACCGCTATGGGTTTGTAGCCAACTTCAGTATGGAGCTACCCAGCTTTG
Haseluenne 2003   CTACCGCTACGGGTTTGTAGCCAATTTCAGTATGGAGCTACCCAGTTTTG
Mexico 2009       TTATCGCTATGGATTTGTGGCTAATTTTAGCATGGAGCTACCCAGCTTTG
                   ** ***** ** ***** ** ** ** ** *********** ** ****

SSIV 18/10 PB1    GGGTGTCTGGAGTTAATGAATCGGCTGACATGAGCATTGGAGTAACAGTG
SSIV 294/09 PB1   GGGTGTCTGGAGTTAATGAATCGGCTGACATGAGCATTGGAGTAACAGTG
SSIV 247/09 PB1   GGGTGTCTGGAGTTAATGAATCAGCTGACATGAGCATTGGAGTGACAGTG
Haseluenne 2003   GGGTATCTGGAGTTAATGAATCGGCTGATATGAGCATCGGAGTGACAGTG
Mexico 2009       GAGTGTCTGGAGTAAATGAATCAGCTGACATGAGTATTGGAGTAACAGTG
                  * ** ******** ******** ***** ***** ** ***** ******

SSIV 18/10 PB1    ATAAAAAACAACATGATAAACAATGATCTCGGGCCAGCAACAGCCCAAAT
SSIV 294/09 PB1   ATAAAAAACAACATGATAAACAATGATCTCGGGCCAGCAACAGCCCAAAT
SSIV 247/09 PB1   ATAAAAAACAACATGATAAACAATGATCTCGGCCCAGCAACAGCCCAAAT
Haseluenne 2003   ATAAAGAACAACATGATAAACAATGATCTCGGGCCAGCAACTGCCCAAAT
Mexico 2009       ATAAAGAACAACATGATAAACAATGACCTTGGACCTGCAACGGCCCAGAT
                  ***** ******************** ** ** ** ***** ***** **

SSIV 18/10 PB1    GGCTCTTCAACTATTCATTAAGGACTACAGGTATACATACCGATGCCACA
SSIV 294/09 PB1   GGCTCTTCAACTATTCATTAAGGACTACAGGTACACATACCGATGCCACA
SSIV 247/09 PB1   GGCTCTTCAACTATTCATTAAGGACTACAGGTATACATACCGATGCCACA
Haseluenne 2003   GGCTCTTCAGCTATTCATTAAGGACTACAGGTATACATACCGGTGCCACA
Mexico 2009       GGCTCTTCAATTGTTCATCAAAGACTACAGATACACATATAGGTGCCATA
                  *********  * ***** ** ******** ** *****  * ***** *

SSIV 18/10 PB1    GAGGTGACACGCAAATCCAAACGAAAAGATCATTTGAGCTGAAAAAACTA
SSIV 294/09 PB1   GAGGTGACACGCAAATCCAAACGAAAAGATCATTTGAGCTGAAAAAACTA
SSIV 247/09 PB1   GAGGTGACACGCAAATCCAAACGAAAAGATCATTCGAGCTGAAAAAACTA
Haseluenne 2003   GAGGTGACACGCAAATCCAAACGAAAAGATCATTCGAGCTAAAAAGACTA
Mexico 2009       GGGGAGACACACAAATTCAGACGAGAAGATCATTTGAGTTAAAGAAGCTG
                  * ** ***** ***** ** **** ********* *** * ** *  **

SSIV 18/10 PB1    TGGGAGCAAACCCATTCAAAGACAGGACTACTGGTTTCTGATGGAGGACC
SSIV 294/09 PB1   TGGGAGCAAACCCATTCAAAGACAGGACTACTGGTTTCTGATGGAGGACC
SSIV 247/09 PB1   TGGGAGCAAACCCATTCAAAGGCAGGACTACTGGTTTCTGATGGAGGACC
Haseluenne 2003   TGGGAGCAAACTCATTCAAAGGCAGGACTACTGATTTCAGATGGAGGTCC
Mexico 2009       TGGGATCAAACCCAATCAAAGGTAGGGCTATTAGTATCAGATGGAGGACC
                  ***** ***** ** ******  *** *** *  * ** ******** **

SSIV 18/10 PB1    AAATCTTTACAATATCCGGAATCTCCACATTCCGGAAGTCTGCTTGAAAT
SSIV 294/09 PB1   AAATCTTTACAATATCCGGAATCTCCACATTCCGGAAGTCTGCTTGAAAT
SSIV 247/09 PB1   AAATCTTTACAATATCCGAAATCTCCACATTCCGGAAGTCTGCTTGAAAT
Haseluenne 2003   AAATCTGTACAATATCCGGAATCTCCACATCCCGGAAGTATGCTTGAAGT
Mexico 2009       AAACTTATACAATATACGGAATCTTCACATTCCTGAAGTCTGCTTAAAAT
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SSIV 18/10 PB1    GGGAGCTAATGGATGTAGATTATCGGGGAAGATTGTGCAATCCTCTGAAT
SSIV 294/09 PB1   GGGAGCTAATGGATGTAGATTATCGGGGAAGATTGTGTAATCCTCTGAAT
SSIV 247/09 PB1   GGGAGCTAATGGATGTAGATTATCGGGGAAGATTGTGTAATCCTCTGAAT
Haseluenne 2003   GGGAGCTGATGGATGTAGATTATCGGGGAAGATTGTGTAATCCTCTGAAT
Mexico 2009       GGGAGCTAATGGATGATGATTATCGGGGAAGACTTTGTAATCCCCTGAAT
                  ******* *******  *************** * ** ***** ******

SSIV 18/10 PB1    CCGTTTGTCAACCATAAGGGAATTGAGTCCGTAAACAGTGCCATGGTAAT
SSIV 294/09 PB1   CCGTTTGTCAACCATAAGGTAATTGAGTCCGTAAACAGTGCCATGGTAAT
SSIV 247/09 PB1   CCGTTTGTCAACCATAAGGGAATTGAGTCTGTAAACAGTGCCATGGTAAT
Haseluenne 2003   CCATTTGTCAACCATAAGGGAATTGAGTCCGTAAACAGTGCTGTGGTAAT
Mexico 2009       CCCTTTGTCAGTCATAAAGAGATTGATTCTGTAAACAATGCTGTGGTAAT
                  ** *******  ***** *  ***** ** ******* ***  *******

SSIV 18/10 PB1    GCCAGCCCATGGTCCAGCCAAAAGCATGGAATATGATGCTGTTGCAACTA
SSIV 294/09 PB1   GCCAGCCCATGGTCCAGCTAAAAGCATGGAATATGATGCTGTTGCAACTA
SSIV 247/09 PB1   GCCAGCCCATGGTCCAGCTAAAAGCATGGAATATGATGCTGTTGCAACTA
Haseluenne 2003   GCCAGCACACGGTCCAGCTAAGAGCATGGAATATGATGCTGTTGCAACTA
Mexico 2009       GCCAGCCCATGGTCCAGCCAAAAGCATGGAATATGATGCCGTTGCAACTA
                  ****** ** ******** ** ***************** **********

SSIV 18/10 PB1    CACATTCTTGGATTCCAAAAAGGAATCGTTCCATTCTCAATACCAGCCAA
SSIV 294/09 PB1   CACATTCTTGGATTCCAAAAAGGAATCGTTCCATTCTCAATACCAGCCAA
SSIV 247/09 PB1   CACATTCTTGGGTTCCAAAAAGGAATCGTTCCATTCTCAATACCAGTCAA
Haseluenne 2003   CACATTCTTGGACTCCAAAAAGGAACCGTTCCATCCTCAATACCAGCCAA
Mexico 2009       CACATTCCTGGATTCCCAAGAGGAATCGTTCTATTCTCAACACAAGCCAA
                  ******* ***  *** ** ***** ***** ** ***** ** ** ***

SSIV 18/10 PB1    AAGGGGATTCTTGAGGATGAACAAATGTACCAGAAATGCTGCAATCTATT
SSIV 294/09 PB1   AGGGGAATTCTTGAGGATGAACAAATGTACCAGAAATGCTGCAATCTATT
SSIV 247/09 PB1   AGGGGAATTCTTGAGGATGAGCAAATGTACCAGAAGTGTTGCAATCTATT
Haseluenne 2003   AGGGGAATTCTTGAGGATGAACAAATGTACCAGAAGTGCTGCAATCTATT
Mexico 2009       AGGGGAATTCTTGAGGATGAACAGATGTACCAGAAGTGCTGCAATCTATT
                  * *** ************** ** *********** ** ***********

SSIV 18/10 PB1    CGAGAAATTCTTCCCTAGCAGTTCATACAGGAGGCCAGTTGGAATTTCAA
SSIV 294/09 PB1   CGAGAAATTCTTCCCTAGCAGTTCATACAGGAGGCCAGTTGGAATTTCAA
SSIV 247/09 PB1   CGAGAAATTCTTCCCTAGCAGTTCATACAGGAGGCCAGTTGGAATTTCAA
Haseluenne 2003   TGAGAAATTCTTCCCTAGCAGTTCATACAGGAGGCCAGTTGGAATTTCCA
Mexico 2009       CGAGAAATTTTTCCCTAGCAGTTCATATAGGAGACCGGTTGGAATTTCTA
                   ******** ***************** ***** ** *********** *

SSIV 18/10 PB1    GCATGGTGGAGGCCATGGTATCTAGGGCCAGAATTGATGCACGGATTGAG
SSIV 294/09 PB1   GCATGGTGGAGGCCATGGTATCTAGGGCCAGAATTGATGCACGGATTGAG
SSIV 247/09 PB1   GCATGGTGGAGGCCATGGTATCTAGGGCCAGAATTGATGCACGGATTGAT
Haseluenne 2003   GCATGGTGGAGGCCATGGTATCTAGGGCCAGAATTGATGCACGGATTGAT
Mexico 2009       GCATGGTGGAGGCCATGGTGTCTAGGGCCCGGATTGATGCCAGGGTCGAC
                  ****************** ********* * ********  ** * **

SSIV 18/10 PB1    TTCGAGTCTGGAAAGATTAAGAAAGAAGAATTTGCTGAGATCATGAAGAT
SSIV 294/09 PB1   TTCGAGTCTGGAAAGATTAAGAAAGAAGAATTTGCTGAGATCATGAAGAT
SSIV 247/09 PB1   TTCGAGTCTGGAAAGATTAAGAAAGAAGAATTTGCTGAGATCATGAAGAT
Haseluenne 2003   TTCGAGTCTGGAAGGATTAAGAAAGAAGAGTTTGCTGAGATCATGAAGAT
Mexico 2009       TTCGAGTCTGGACGGATCAAGAAAGAAGAGTTCTCTGAGATCATGAAGAT
                  ************  *** *********** **  ****************

SSIV 18/10 PB1    CTGTTCCACCATTGAAGAGTTCAAACGGCAAAAGTAGTGAATTTAGCTTG
SSIV 294/09 PB1   CTGTTCCACCATTGAAGAGTTCAAACGGCAAAAGTAGTGAATTTAGCTTG
SSIV 247/09 PB1   CTGTTCCACCATTGAAGAGTTCAAACGGCAAAAGTAGTGAATTTAGCTTG
Haseluenne 2003   CTGTTCCACCATTGAAGAGCTCGGACGGCAAAAGTAGTGAATTTGGCTTG
Mexico 2009       CTGTTCCACCATTGAAGAACTCAGACGGCAAAAATAATGAATTTAACTTG
                  ******************  **  ********* ** *******  ****

SSIV 18/10 PB1    TCCTTCATGAAAAAATGCCTTGTTTCTACTAATACGAGACGATATAAGGG
SSIV 294/09 PB1   TCCTTCATGAAAAAATGCCTTGTTTNTANTAATACGAGACGATATAAGGG
SSIV 247/09 PB1   TCCTTCAN-AAAAAATGCCTTGTTTCTACTAATACGAGAC----------
Haseluenne 2003   TCCTTCATGAAAAAATGCCTTGTTTCTACT--------------------
Mexico 2009       TCCTTCATGAAA---------------------------------------
                  *******  ***

SSIV 18/10 PB1    CGAATTCCAGCACACTGGCGGCCGTTACTAG--------
SSIV 294/09 PB1   CGAATTCCAGCACACTGGCGGCCGTTACTAGNNNCCGAG
SSIV 247/09 PB1   ----------------------------------------
Haseluenne 2003   ----------------------------------------
Mexico 2009       ----------------------------------------
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Legend:
Nucleotide sequences of segment 3.

Appendix 7b

PB1 protein

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SSIV 294/09 PB1      MDVNPTLLFLKVPAQNAISTTFPYTGDPPYSHGTGTGYTMDTVNRTHQYS
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SSIV 18/10 PB1        MDVNPTLLFLKVPAQNAISTTFPYTGDPPYSHGTGTGYTMDTVNRTHQYS
SSIV 247/09 PB1       MDVNPILLFLKVPAQNAISTTFPYTGDPPYSHGTGTGYTMDTVNRTHQYS
Haseluenne 2003       MDVNPTLLFLKVPAQNAISTTFPYTGDPPYSHGTGTGYTMDTVNRTHQYS
Mexico 2009           MDVNPTLLFLKIPAQNAISTTFPYTGDPPYSHGTGTGYTMDTVNRTHQYS
                      ***** *****:**************************************

SSIV 294/09 PB1       ERGRWTTNTETGAPQLNPIDGPLPEDNEPSGYAQTDCVLEAMAFLEESHP
SSIV 18/10 PB1        ERGRWTTNTETGAPQLNPIDGPLPEDNEPSGYAQTDCVLEAMAFLEESHP
SSIV 247/09 PB1       ERGKWTTNTETGAPQLNPIDGPLPEDNEPSGYAQTDCVLEAMAFLEESHP
Haseluenne 2003       ERGRWTTNTETGAPQLNPIDGPLPEDNEPSGYAQTDCVLEAMAYLEESHP
Mexico 2009           EKGKWTTNTETGAPQLNPIDGPLPEDNEPSGYAQTDCVLEAMAFLEESHP
                      *:*:*****************************************:******

SSIV 294/09 PB1       GIFENSCLETMEVVQQTRVDKLTQGRQTYDWTLNRNQPAATALANTIEVF
SSIV 18/10 PB1        GIFENSCLETMEVVQQTRVDKLTQGRQTYGWTLNRNQPAATALANTIEVF
SSIV 247/09 PB1       GIFENSCLETIEVVQQTRVDKLTQGRQTYDWTLNRNQPAATALANTIEVF
Haseluenne 2003       GIFENSCLETMEVVQQTRVDKLTQGRQTYDWTLNRNQPAATALANTIEVF
Mexico 2009           GIFENSCLETMEVVQQTRVDKLTQGRQTYDWTLNRNQPAATALANTIEVF
                      **********:******************.********************

SSIV 294/09 PB1       RMNSLTANESGRLIDFLKDVMESMDKEEMEITTHFQRKRRIRDNMTKKMV
SSIV 18/10 PB1        RMNSLTANESRRLIDFLKDVMESMDKEEMEITTHFQRKRRIRDNMTKKMV
SSIV 247/09 PB1       RMNSLTANESGRLIDFLKDVMESMDKEEMEITTHFQRKRRIRDNMTKKMV
Haseluenne 2003       RLNSLTANESGRLIDFLKDVMESMDKEEMEITTHFQRKRRIRDNMTKKMI
Mexico 2009           RSNGLTANESGRLIDFLKDVMESMNKEEIEITTHFQRKRRVRDNMTKKMV
                      * *.****** *************:***:***********:********:

SSIV 294/09 PB1       TQRTIGKKKQKLNKKNYLIRALTLNTMTKDAERGKLKRRAIATPGVQIRG
SSIV 18/10 PB1        TQRTIGKKKQKLNKKNYLIRALTLNTMTKDAERGKLKRRAIATPGMQIRG
SSIV 247/09 PB1       TQRTIGKKKQKLNKKNYLIRALTLNTMTKDAERGKLKRRAIATPGMQIRG
Haseluenne 2003       TQRTIGKKKQKLNKRSYLIRALTLNTMTKDAERGKLKRRAIATPGMQIRG
Mexico 2009           TQRTIGKKKQRLNKRGYLIRALTLNTMTKDAERGKLKRRAIATPGMQIRG
                      **********:***:.*****************************:****

SSIV 294/09 PB1       FVYFVETLARSICEKLEQSGLPVGGNEKKAKLANVVRKMMTNSQDTELSF
SSIV 18/10 PB1        FVYFVETLARSICEKLEQSGLPVGGNEKKAKLANVARKMMTNSQDTELSF
SSIV 247/09 PB1       FVYFVETLARSICEKLEQSGLPVGGNEKKAKLANVVRKMMTNSQDTELSF
Haseluenne 2003       FVYFVETLARSICEKLEQSGLPVGGNEKKAKLANVVRKMMTNSQDTELSF
Mexico 2009           FVYFVETLARSICEKLEQSGLPVGGNEKKAKLANVVRKMMTNSQDTEISF
                      **********************************.***********:**

SSIV 294/09 PB1       TITGDNTKWNENQNPRMFLAMITYITRNQPEWFRNVLSIAPIMFSNKMAR
SSIV 18/10 PB1        TITGDNTKWNENQNPRMFLAMITYVTRNQPEWFRNVLSIAPIMFSNKMAR
SSIV 247/09 PB1       TITGDNTKWNENQNPRMFLAMITYITRNQPEWFRNVLSIAPIMFSNKMAR
Haseluenne 2003       TITGDNTKWNENQNPRVFLAMITYITRNQPEWFRNVLSIAPIMFSNKMAR
Mexico 2009           TITGDNTKWNENQNPRMFLAMITYITRNQPEWFRNILSMAPIMFSNKMAR
                      ****************:*******:**********:**:***********

SSIV 294/09 PB1       LGKGYMFESKSMKLRTQIPAEMLADIDLKYFNESTRKKIEKIRPLLIDGT
SSIV 18/10 PB1        LGKGYMFESKSMKLRAQIPAEMLADIDLKYFNGSTRKKIEKIRPLLIDGT
SSIV 247/09 PB1       LGKGYMFESKSMKLRTQIPAEMLADIDLKYFNESTRKKIEKIRPLLIDGT
Haseluenne 2003       LGKGYMFESKSMKLRTQIPAEMLADIDLKYFNESTRKKIEKIRPLLINGT
Mexico 2009           LGKGYMFESKRMKIRTQIPAEMLASIDLKYFNESTKKKIEKIRPLLIDGT
                      ********** **:*:********.******* **:***********:**

SSIV 294/09 PB1       ASLSPGMMMGMFNMLSTVLGVSILNLGQKRYTKTTYWWDGLQSSDDFALI
SSIV 18/10 PB1        ASLSPGMMMGMFNMLSTVLGVSILNLGQKRYTKTTYWWDGLQSSDDFALI
SSIV 247/09 PB1       ASLSPGMMMGMFNMLSTVLGVSILNLGQKRYTKTTYWWDGLQSSDDFALI
Haseluenne 2003       ASLSPGMMMGMFNMLSTVLGVSILNLGQKRYTKTTYWWDGLQSSDDFALI
Mexico 2009           ASLSPGMMMGMFNMLSTVLGVSILNLGQKKYTKTIYWWDGLQSSDDFALI
                      *****************************:**** ***************

SSIV 294/09 PB1       VNAPNHEGIQAGVDRFYRTCKLVGINMSKKKSYINRTGTFEFTSFFYRYG
SSIV 18/10 PB1        VNAPNHEGIQAGVDRFYRTCKLVGINMSKKKSYINRTGTFEFTSFFYRYG
SSIV 247/09 PB1       VNAPNHEGIQAGVDRFYRTCKLVGINMSKKKSYINRTGTFEFTSFFYRYG
Haseluenne 2003       VNAPSHEGIQAGVDRFYRTCKLVGINMSKKKSYINRTGTFEFTSFFYRYG
Mexico 2009           VNAPNHEGIQAGVDRFYRTCKLVGINMSKKKSYINKTGTFEFTSFFYRYG
                      ****.*****************************:***************

SSIV 294/09 PB1       FVANFSMELPSFGVSGVNESADMSIGVTVIKNNMINNDLGPATAQMALQL
SSIV 18/10 PB1        FVANFSMELPSFGVSGVNESADMSIGVTVIKNNMINNDLGPATAQMALQL
SSIV 247/09 PB1       FVANFSMELPSFGVSGVNESADMSIGVTVIKNNMINNDLGPATAQMALQL
Haseluenne 2003       FVANFSMELPSFGVSGVNESADMSIGVTVIKNNMINNDLGPATAQMALQL
Mexico 2009           FVANFSMELPSFGVSGVNESADMSIGVTVIKNNMINNDLGPATAQMALQL
                      **************************************************

SSIV 294/09 PB1       FIKDYRYTYRCHRGDTQIQTKRSFELKKLWEQTHSKTGLLVSDGGPNLYN
SSIV 18/10 PB1        FIKDYRYTYRCHRGDTQIQTKRSFELKKLWEQTHSKTGLLVSDGGPNLYN
SSIV 247/09 PB1       FIKDYRYTYRCHRGDTQIQTKRSFELKKLWEQTHSKAGLLVSDGGPNLYN
Haseluenne 2003       FIKDYRYTYRCHRGDTQIQTKRSFELKRLWEQTHSKAGLLISDGGPNLYN
Mexico 2009           FIKDYRYTYRCHRGDTQIQTRRSFELKKLWDQTQSKVGLLVSDGGPNLYN
                      ********************:******:**:**:**.***:*********

SSIV 294/09 PB1       IRNLHIPEVCLKWELMDVDYRGRLCNPLNPFVNHKVIESVNSAMVMPAHG
SSIV 18/10 PB1        IRNLHIPEVCLKWELMDVDYRGRLCNPLNPFVNHKGIESVNSAMVMPAHG
SSIV 247/09 PB1       IRNLHIPEVCLKWELMDVDYRGRLCNPLNPFVNHKGIESVNSAMVMPAHG
Haseluenne 2003       IRNLHIPEVCLKWELMDVDYRGRLCNPLNPFVNHKGIESVNSAVVMPAHG
Mexico 2009           IRNLHIPEVCLKWELMDDDYRGRLCNPLNPFVSHKEIDSVNNAVVMPAHG
                      ***************** **************.** *:***.*:******
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SSIV 294/09 PB1      PAKSMEYDAVATTHSWIPKRNRSILNTSQRGILEDEQMYQKCCNLFEKFF
SSIV 18/10 PB1       PAKSMEYDAVATTHSWIPKRNRSILNTSQKGILEDEQMYQKCCNLFEKFF
SSIV 247/09 PB1      PAKSMEYDAVATTHSWVPKRNRSILNTSQRGILEDEQMYQKCCNLFEKFF
Haseluenne 2003      PAKSMEYDAVATTHSWTPKRNRSILNTSQRGILEDEQMYQKCCNLFEKFF
Mexico 2009          PAKSMEYDAVATTHSWIPKRNRSILNTSQRGILEDEQMYQKCCNLFEKFF
                     *************** ************:********************

SSIV 294/09 PB1      PSSSYRRPVGISSMVEAMVSRARIDARIEFESGKIKKEEFAEIMKICSTI
SSIV 18/10 PB1       PSSSYRRPVGISSMVEAMVSRARIDARIEFESGKIKKEEFAEIMKICSTI
SSIV 247/09 PB1      PSSSYRRPVGISSMVEAMVSRARIDARIDFESGKIKKEEFAEIMKICSTI
Haseluenne 2003      PSSSYRRPVGISSMVEAMVSRARIDARIDFESGRIKKEEFAEIMKICSTI
Mexico 2009          PSSSYRRPVGISSMVEAMVSRARIDARVDFESGRIKKEEFSEIMKICSTI
                     ****************************::****:******:*********

SSIV 294/09 PB1      EEFKRQK
SSIV 18/10 PB1       EEFKRQK
SSIV 247/09 PB1      EEFKRQK
Haseluenne 2003      EELGRQK
Mexico 2009          EELRRQK
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Legend:
Amino acid sequences of PB1 protein.

Appendix 8a

Segment 1

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SSIV 294/09 PB2      AGCGAAAGCAGGTCAAATATATTCAATATGGAGAGAATAAAAGAGTTAAG
SSIV 18/10 PB2       AGCGAAAGCAGGTCAAATATATTCAATATGGAGAGAATAAAAGGGTTAAG
Haseluenne 2003      AGCGAAAGCAGGTCAAATATATTCAATATGGAGAGAATAAAAGAATTAAG
Mexico 2009          -------------------------TATGGAGAGAATAAAAGAACTGAG
                                               ****************   * **

SSIV 294/09 PB2      AGATCTGATGTCGCAGTCTCGCACTCGCGAGATACTGACAAAGACCACTG
SSIV 18/10 PB2       AGATCTGATGTCGCAATCTCGCACTCGCGAGATACTGACAAAGACCACTG
Haseluenne 2003      AGATCTGATGTCGCAGTCTCGCACTCGCGAGATACTGACAAAAACCACTG
Mexico 2009          AGATCTAATGTCGCAGTCCCGCACTCGCGAGATACTCACTAAGACCACTG
                     ****** ******** ** ***************** ** ** *******

SSIV 294/09 PB2      TGGACCATATGGCAATAATAAAAAAATACACATCAGGAAGACAAGAGAAG
SSIV 18/10 PB2       TGGACCATATGGCAATAATAAAAAAATACACATCAGGAAGACAAGAGAAG
Haseluenne 2003      TAGACCATATGGCAATAATAAAAAAATACACATCAGGGAGACAAGAGAAG
Mexico 2009          TGGACCATATGGCCATAATCAAAAAGTACACATCAGGAAGGCAAGAGAAG
                     * *********** ***** ***** *********** ** *********

SSIV 294/09 PB2      AACCCCTCTCTCAGAATGAAATGGATGATGGCAATGAAGTATCCGATTAC
SSIV 18/10 PB2       AACCCCTCTCTCAGAATGAAATGGATGATGGCAATGAAGTATCCGATTAC
Haseluenne 2003      AACCCCGCTCTCAGAATGAAATGGATGATGGCAATGAAATATCCAATTAC
Mexico 2009          AACCCCGCACTCAGAATGAAGTGGATGATGGCAATGAGATACCCAATTAC
                     ****** * *********** ****************  ** ** *****

SSIV 294/09 PB2      AGCAGACAGGAGGATAATGGAGATGATTCCTGAAAGAAGTGAACAAGGAC
SSIV 18/10 PB2       AGCAGACAGGAGGATAATGGAGATGATTCCTGAAAGAAATGAACAAGGAC
Haseluenne 2003      AGCAGACAGGAGGATAATGGAGATGATTCCTGAAAGAAATGAACAAGGAC
Mexico 2009          AGCAGACAAGAGAATAATGGACATGATTCCAGAGAGGAATGAACAAGGAC
                     ******** *** ******** ******** ** ** * ***********

SSIV 294/09 PB2      AAATACTTTGGAGTAAGACAAATGATGCTGGATCAGATAGGGTGATGGTG
SSIV 18/10 PB2       AAATACTTTGGAGTAAGACAAATGATGCTGGATCAGATAGGGTGATGGTG
Haseluenne 2003      AAATACTTTGGAGTAAGACAAATGATGCTGGATCAGATAGGGTGATGGTA
Mexico 2009          AAACCCTCTGGAGCAAAACAAACGATGCTGGATCAGACCGAGTGATGGTA
                     ***  ** ***** ** ***** **************  * ********

SSIV 294/09 PB2      TCACCCCTAGCCGTAACTTGGTGGAATAGGAATGGACCGACAACAGATAC
SSIV 18/10 PB2       TCACCCCTAGCCGTAACTTGGTGGAATAGGAATGGACCGACAGCAGATAC
Haseluenne 2003      TCACCCCTAGCCGTAACTTGGTGGAATAGGAATGGACCGACAACAGATAC
Mexico 2009          TCACCTCTGGCCGTAACATGGTGGAATAGGAATGGCCCAACAACAAGTAC
                     ***** ** ******** ***************** ** *** **  ***

SSIV 294/09 PB2      AGTCCACTATCCTAAAGTATACAAAACATATTTTGAAAAAGTTGAAAGGT
SSIV 18/10 PB2       AGTCCACTATCCTAAAGTATACAAAACATATTTTGAAAAAGTAGAAAGGT
Haseluenne 2003      AGTCCACTATCCTAAAGTATACAAAACATATTTTGAGAAAGTTGAAAGGT
Mexico 2009          AGTTCATTACCCTAAGGTATATAAAACTTATTTCGAAAAGGTCGAAAGGT
                     *** ** ** ***** ***** ***** ***** ** ** ** *******

SSIV 294/09 PB2      TGAAGCATGGGACCTTTGGTCCCGTCCATTTTCGAAATCAAGTTAAAATA
SSIV 18/10 PB2       TGAAGCATGGGACCTTTGGTCCCGTCCATTTTCGAAATCAAGTTAAAATA
Haseluenne 2003      TGAAGCATGGGACCTTTGGTCCCGTCCATTTTCGAAATCAAGTTAAAATA
Mexico 2009          TGAAACATGGTACCTTCGGCCCTGTCCACTTCAGAAATCAAGTTAAAATA
                     **** ***** ***** ** ** ***** **  *****************

SSIV 294/09 PB2      CGCCGAAGGGTTGACATAAACCCAGGCCATGCAGACCTCAATGCCAAAGA
SSIV 18/10 PB2       CGCCGAAGGGTTGACATAAACCCAGGCCATGCAGACCTCAATGCCAAGGA
Haseluenne 2003      CGCCGAAGAGTTGACATAAACCCAGGCCATGCAGATCTCAGTGCCAAGGA
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Mexico 2009          AGGAGGAGAGTTGATACAAACCCTGGCCATGCAGATCTCAGTGCCAAGGA
                      *  * ** ***** * ****** ********** **** ****** **

SSIV 294/09 PB2      AGCACAAGATGTTATCATGGAAGTTGTTTTCCCAAATGAAGTGGGAGCCA
SSIV 18/10 PB2       AGCACAAGATGTTATCATGGAAGTTGTTTTCCCAAATGAAGTGGGAGCCA
Haseluenne 2003      TGCACAAGATGTTATCATGGAAGTTGTTTTCCCAAATGAAGTGGGAGCCA
Mexico 2009          GGCACAGGATGTGATTATGGAAGTTGTTTTCCCAAATGAAGTGGGGGCAA
                      ***** ***** ** ***************************** ** *

SSIV 294/09 PB2      GAATACTAACATCAGAGTCACAATTAACAATAACAAAAGAGAAGAAAGAG
SSIV 18/10 PB2       GAATACTAACATCAGAGTCACAATTAACAATAACAAAAGAGAAGAAAGAG
Haseluenne 2003      GAATACTAACATCAGAGTCACAATTAACAATAACAAAAGAGAAGAAAGAG
Mexico 2009          GAATACTGACATCAGAGTCACAGCTGGCAATAACAAAAGAGAAGAAAGAA
                     ******* **************  *  **********************

SSIV 294/09 PB2      GAACTCCAAGATTGTAAGATTGCCCCTTTAATGGTGGCATACATGTTGGA
SSIV 18/10 PB2       GAACTCCAAGATTGTAAGATTGCCCCTTTAATGGTGGCATACATGTTGGA
Haseluenne 2003      GAGCTCCAGGATTGTAAGATTGCCCCTTTAATGGTGGCATACATGTTGGA
Mexico 2009          GAGCTCCAGGATTGTAAAATTGCTCCCTTGATGGTGGCGTACATGCTAGA
                     ** ***** ******** ***** ** ** ******** ****** * **

SSIV 294/09 PB2      AAGAGAACTGGTCCGCAAAACCAGATTCCTGCCGGTGGCAGGTGGAACAA
SSIV 18/10 PB2       AAGAGAACTGGTCCGCAAAACCAGATTCCTGCCGGTGGCAGGTGGAACAA
Haseluenne 2003      AAGAGAACTGGTCCGCAAAACCAGATTCCTGCCGGTAGCAGGTGGAACAA
Mexico 2009          AAGAGAATTGGTCCGTAAAACAAGGTTTCTCCCAGTAGCCGGCGGAACAG
                     ******* ******* ***** ** ** ** ** ** ** ** ******

SSIV 294/09 PB2      GCAGTGTCTACATTGAAGTATTGCACTTAACTCAAGGAACCTGCTGGGAG
SSIV 18/10 PB2       GCAGTGTCTACATTGAAGTATTGCACTTAACTCAAGGAACCTGCTGGGAA
Haseluenne 2003      GCAGTGTCTACATTGAAGTATTGCACTTAACTCAAGGAACCTGCTGGGAA
Mexico 2009          GCAGTGTTTATATTGAAGTGTTGCACTTAACCCAAGGGACGTGCTGGGAG
                     ******* ** ******** ********** ***** ** ********

SSIV 294/09 PB2      CAGATGTACACTCCAGGTGGAGAGGTGAAGAATGATGATATAGACCAGAG
SSIV 18/10 PB2       CAGATGTACACTCCGGGTGGAGAGGTAAAGAATGATGATATAGACCAGAG
Haseluenne 2003      CAGATGTACACTCCAGGTGGAAAGGTGAAGAATGATGATGTAGACCAGAG
Mexico 2009          CAGATGTACACTCCAGGAGGAGAAGTGAGAAATGATGATGTTGACCAAAG
                     ************** ** *** * ** *  ********* * ***** **

SSIV 294/09 PB2      TTTGATCATTGCTGCCAGAAACATTGTTAGAAGAGCAATAGTGTCAGCAG
SSIV 18/10 PB2       TTTGATCATTGCTGCCAGAAACATTGTTAGAAGAGCAATAGTGTCAGCAG
Haseluenne 2003      TTTGATCATTGCTGCCAGAAACATTGTCAGAAGAGCAATAGTATCAGCAG
Mexico 2009          TTTGATTATCGCTGCTAGAAACATAGTAAGAAGAGCAGCAGTGTCAGCAG
                     ****** ** ***** ******** ** *********  *** *******

SSIV 294/09 PB2      ATCCATTGGCATCACTATTGGAGATGTGTCACGGCACACAAATTGGTGGG
SSIV 18/10 PB2       ATCCATTGGCATCACTATTGGAGATGTGTCACGGCACACAAATTGGTGGG
Haseluenne 2003      ATCCACTGGCATCACTATTAGAGATGTGTCACAGCACACAAATTGGTGGG
Mexico 2009          ACCCATTAGCATCTCTCTTGGAAATGTGCCACAGCACACAGATTGGAGGA
                     * *** * ***** ** ** ** ***** *** ******* ***** **

SSIV 294/09 PB2      ATAAGGATGGTTGACATCCTTAAACAAAATCCAACAGAAGAGCAAGCCGT
SSIV 18/10 PB2       ATAAGGATGGTTGACATCCTTAAACAAAATCCTACGGAAGAGCAAGCTGT
Haseluenne 2003      ATAAGGATGGTTGACATCCTTAAACAAAATCCAACGGAAGAGCAAGCCGT
Mexico 2009          GTAAGGATGGTGGACATCCTTAGACAGAATCCAACTGAGGAACAAGCCGT
                     ********** ********** *** ***** ** ** ** ***** **

SSIV 294/09 PB2      GGATATATGCAAAGCAGCAATGGGTTTGAGAATCAGCTCCTCCTTTAGCT
SSIV 18/10 PB2       GGATATATGCAAAGCAGCAATGGGTTTGAGAATCAGCTCCTCCTTTAGCT
Haseluenne 2003      AGATATATGCAAAGCAGCAATGGGTTTGAGAATCAGCTCCTCCTTTAGCT
Mexico 2009          AGACATATGCAAGGCAGCAATAGGGTTGAGGATTAGCTCATCTTTCAGTT
                     ** ******** ******** ** ***** ** ***** ** ** ** *

SSIV 294/09 PB2      TTGGAGGTTTCACTTTCAAAAGAACAAGTGGATCATCTGTTAAAAGGGAA
SSIV 18/10 PB2       TTGGAGGTCTCACTTTCAAAAGAACAAGTGGATCATCTGTTAAAAAGGAA
Haseluenne 2003      TTGGAGGTTTCACTTTCAAAAGAACAAGTGGATCATCTGTTAAAAGGGAA
Mexico 2009          TTGGTGGGTTCACTTTCAAAAGGACAAGCGGATCATCAGTCAAGAAAGAA
                     **** **  ************** ***** ******** ** ** *  ***

SSIV 294/09 PB2      GAAGAAGTGCTTACAGGCAATCTCCAAGCACTGAAGATAAGGGTACATGA
SSIV 18/10 PB2       GAAGAAGTGCTTACAGGCAATCTCCAAGCACTGAAGATAAGGGTACATGA
Haseluenne 2003      GAAGAAGTGCTCACAGGCAATCTCCAAGCACTGAAGATAAGGGTACATGA
Mexico 2009          GAAGAAGTGCTAACGGGCAACCTCCAAACACTGAAAATAAGAGTACATGA
                     *********** ** ***** ****** ******* ***** ********

SSIV 294/09 PB2      GGGGTATGAGGAATTTACAATGGTTGGAAGAAGAGCGACAGCCATCTTAA
SSIV 18/10 PB2       GGGGTATGAGGAATTTACAATGGTTGGAAGAAGAGCAACAGCCATCTTAA
Haseluenne 2003      GGGGTATGAGGAGTTCACAATGGTTGGGAAACGAGCAACAGCCATCTTAA
Mexico 2009          AGGGTATGAAGAATTCACAATGGTTGGGAGAAGAGCAACAGCTATTCTCA
                     ******** ** ** *********** * * **** ***** **  * *

SSIV 294/09 PB2      GGAAAGCAACTAGAAGGCTGATTCAGCTGATAGTAAGTGGAAGAGACGAA
SSIV 18/10 PB2       GGAAAGCAACTAGAAGGCTGATTCAGCTGATAGTAAGTGGAAGAGACGAA
Haseluenne 2003      GGAAAGCAACTAGAAGGCTGATTCAGCTGATAGTAAGTGGAAGAGACGAA
Mexico 2009          GAAAGGCAACCAGGAGATTGATCCAGTTGATAGTAAGCGGGAGAGACGAG
                     * ** ***** ** **  **** *** ********** ** ********

SSIV 294/09 PB2      CAGTCAATTGCTGAAGCAATCATAGTAGCAATGGTGTTCTCACAAGAAGA
SSIV 18/10 PB2       CAGTCAATTGCTGAAGCAATCATAGTAGCAATGGTGTTCTCACAAGAAGA
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Haseluenne 2003      CAGTCAATTGCTGAAGCGATCATAGTAGCAATGGTGTTCTCACAAGAGGA
Mexico 2009          CAGTCAATTGCTGAGGCAATAATTGTGGCCATGGTATTCTCACAAGAGGA
                     ************** ** ** ** ** ** ***** ********** **

SSIV 294/09 PB2      TTGTATGATAAAGGCTGTTCGAGGTGACCTAAATTTTGTAAACAGAGCAA
SSIV 18/10 PB2       TTGTATGATAAAGGCTGTTCGAGGTGATCTAAATTTTGTAAACAGAGCAA
Haseluenne 2003      TTGTATGATAAAGGCTGTCCGAGGTGATCTAAATTTTGTAAACAGAGCAA
Mexico 2009          TTGCATGATCAAGGCAGTTAGGGGCGATCTGAACTTTGTCAATAGGGCAA
                     *** ***** ***** **  * ** ** ** ** ***** ** ** ****

SSIV 294/09 PB2      ACCAAAGGCTGAATCCCATGCATCAACTCCTGAGACACTTCCAAAAGGAT
SSIV 18/10 PB2       ACCAAAGGCTGAATCCCATGCATCAACTCCTAAGACACTTCCAAAAGGAT
Haseluenne 2003      ACCAACGGCTGAATCCCATGCATCAACTCCTGAGACACTTCCAGAAGAAT
Mexico 2009          ACCAGCGACTGAACCCCATGCACCAACTCTTGAGGCATTTCCAAAAAGAT
                     ****  * ***** ******** ****** * ** ** ***** **  **

SSIV 294/09 PB2      GCAAAAGTACTGTTTCAAAATTGGGGAATTGAACCTATTGATAATATCAT
SSIV 18/10 PB2       GCAAAAGTACTGTTTCAAAATTGGGGAATTGAACCTATTGATAATATTAT
Haseluenne 2003      GCAAAAGTGCTGTTTCAAAATTGGGGAATTGAACCTATTGATAATATCAT
Mexico 2009          GCAAAAGTGCTTTTCCAGAACTGGGGAATTGAATCCATCGACAATGTGAT
                     ******** ** ** ** ** ************ * ** ** *** * **

SSIV 294/09 PB2      GGGAATGATAGGTATATTACCCGATATGACTCCAAGCACAGAGATGTCAT
SSIV 18/10 PB2       GGGAATGATAGGTATATTACCCGATATGACACCAAGCACAGAGATGTCAT
Haseluenne 2003      GGGAATGATAGGTATATTACCTGATATGACTCCAAGCACAGAGATGTCAT
Mexico 2009          GGGAATGATCGGAATACTGCCCGACATGACCCCAAGCACGGAGATGTCGC
                     ********* ** *** * ** ** ***** ******** ********

SSIV 294/09 PB2      TAAGAGGAGTGAGGATCAGTAAAACAGGAGTAGACGAATATTCCAGTACT
SSIV 18/10 PB2       TAAGAGGAGTGAGGATCAGTAAAACAGGAGTAGACGAATATTCCAGTACT
Haseluenne 2003      TAAGAGGAGTGAGGGTCAGTAAAACAGGAGTAGACGAATATTCCAGTACT
Mexico 2009          TGAGAGGGATAAGAGTCAGCAAAATGGGAGTAGATGAATACTCCAGCACG
                     * *****  * **  **** ****  ******** ***** ***** **

SSIV 294/09 PB2      GAGAGAGTGGTTGTGAGTATTGATCGTTTCTTGAGAGTTCGAGATCAGCG
SSIV 18/10 PB2       GAGAGAGTGGTTGTGAGTATTGATCGTTTCTTGAGAGTTCGAGATCAGCG
Haseluenne 2003      GAGAGAGTGGTTGTGAGTATTGATCGCTTCTTGAGAGTTCGAGATCAGCG
Mexico 2009          GAGAGAGTGGTAGTGAGTATTGACCGATTTTTAAGGGTTAGAGATCAAAG
                     *********** *********** ** ** ** ** *** *******  *

SSIV 294/09 PB2      GGGGAATGTACTACTATCTCCTGAGGAGGTTAGTGAAACACAAGGGACAG
SSIV 18/10 PB2       GGGAAATGTACTACTGTCTCCTGAGGAGGTTAGCGAAACACAAGGGACAG
Haseluenne 2003      GGGAAATGTACTACTATCTCCTGAGGAGGTTAGCGAAACACAGGGGACAG
Mexico 2009          AGGGAACGTACTATTGTCTCCCGAAGAAGTCAGTGAAACGCAAGGAACTG
                     ** ** ****** * ***** ** ** ** ** ***** ** ** ** *

SSIV 294/09 PB2      AGAAATTGACAATAACCTATTCATCATCAATGATGTGGGAGATCAATGGA
SSIV 18/10 PB2       AGAAATTGACAATAACCTATTCATCATCAATGATGTGGGAGATCAATGGA
Haseluenne 2003      AGAAATTGACAATAACCTATTCATCATCAATGATGTGGGAGATCAATGGA
Mexico 2009          AGAGGTTGACAATAACTTATTCGTCATCAATGATGTGGGAGATCAATGGC
                     ***  *********** ***** *************************

SSIV 294/09 PB2      CCAGAATCAGTGCTCGTTAACACGTATCAATGGATCATTAGAAATTGGGA
SSIV 18/10 PB2       CCAGAATCAGTGCTCGTTAACACGTATCAATGGATCATTAGAAATTGGGA
Haseluenne 2003      CCTGAGTCAGTGCTCATTAACACGTATCAATGGATCATTAGAAATTGGGA
Mexico 2009          CCTGAGTCAGTGCTAGTCAACACTTATCAATGGATAATCAGGAACTGGGA
                     ** ** ********  * ***** *********** ** ** ** *****

SSIV 294/09 PB2      AAATGTGAAGATCCAGTGGTCTCAAGACCCCACAATGCTATATAACAAAA
SSIV 18/10 PB2       AAATGTGAAGATCCAGTGGTCTCAAGACCCCACAATGCTATATAACAAGA
Haseluenne 2003      AACTGTGAAGATCCAATGGTCCCAAGACCCTACAATGCTATACAACAAGA
Mexico 2009          AATTGTGAAAATTCAATGGTCACAAGATCCCACAATGTTATACAACAAAA
                     ** ****** ** ** ***** ***** ** ****** **** ***** *

SSIV 294/09 PB2      TGGAATTTGAGCCATTTCAGTCTCTAATACCTAAAGCAGCCAGAGGTCAA
SSIV 18/10 PB2       TGGAATTTGAGCCATTTCAGTCTCTAATACCTAAAGCAGCCAGAGGTCAA
Haseluenne 2003      TGGAATTTGAGCCATTTCAGTCTCTAGTACCTAAAGCAGCCAGAGGTCAA
Mexico 2009          TGGAATTTGAACCATTTCAGTCTCTTGTCCCTAAGGCAACCAGAAGCCGG
                     ********** **************  * ***** *** ***** * *

SSIV 294/09 PB2      TACAGCGGGTTTGTGAGAACACTATTCCAGCAAATGCGCGATGTACTGGG
SSIV 18/10 PB2       TACAGCGGGTTTGTGAGAACACTATTCCAGCAAATGCGCGATGTACTGGG
Haseluenne 2003      TACAGCGGGTTTGTGAGAACACTATTCCAGCAAATGCGTGATGTGCTGGG
Mexico 2009          TACAGTGGATTCGTAAGGACACTGTTCCAGCAAATGCGGGATGTGCTTGG
                     ***** ** ** ** ** ***** ************** ***** ** **

SSIV 294/09 PB2      GACATTTGATACTGCCCAAATAATAAAGCTACTGCCATTTGCAGCAGCTC
SSIV 18/10 PB2       GACATTTGATACTGCCCAAATAATAAAGCTACTGCCATTTGCAGCAGCTC
Haseluenne 2003      GACATTTGATACTGCCCAAATAATAAAGCTGCTGCCATTTGCAGCAGCTC
Mexico 2009          GACATTTGACACTGTCCAAATAATAAAACTTCTCCCCTTTGCTGCTGCTC
                     ********* **** ************ ** ** ** ***** ** ****

SSIV 294/09 PB2      CACCAGAGCAGAGTAGGATGCAATTCTCTTCTCTAACTGTAAATGTAAGA
SSIV 18/10 PB2       CACCAGAGCAGAGTAGGATGCAATTCTCTTCTCTAACTGTAAATGTAAGA
Haseluenne 2003      CACCAGAGCAGAGTAGGATGCAGTTCTCTTCTCTAACTGTAAACGTAAGA
Mexico 2009          CACCAGAACAGAGTAGGATGCAATTTTCCTCATTGACTGTGAATGTGAGA
                     ******* ************** ** ** **  * ***** ** ** ***

SSIV 294/09 PB2      GGATCAGGACTGAGAATACTCGTAAGAGGTAACTCCCCAGTATTTAACTA
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SSIV 18/10 PB2       GGATCAGGACTGAGAATACTCATAAGAGGTAACTCCCCAGTATTTAACTA
Haseluenne 2003      GGATCAGGACTGAGAATACTCATAAGAGGTAACTCCCCAGTATTTAATTA
Mexico 2009          GGATCAGGGTTGAGGATACTGGTAAGAGGCAATTCTCCAGTATTCAATTA
                     ********  **** *****  ******* ** ** ******** ** **

SSIV 294/09 PB2      CAATAAGACAACAAGAAGGCTAACGGTCCTCGGAAAAGACGCAGGTGCAT
SSIV 18/10 PB2       CAATAAGACAACAAAAAGGCTAACGGTCCTCGGAAAAGACGCAGGTGCGT
Haseluenne 2003      CAATAAGGCAACCAAAAGGCTAACGGTCCTCGGAAAGGACGCAGGTGCAT
Mexico 2009          CAACAAGGCAACCAAACGACTTACAGTTCTTGGAAAGGATGCAGGTGCAT
                     *** *** **** * * * ** ** ** ** ***** ** ******** *

SSIV 294/09 PB2      TAACAGAAGATCCAGATGAGGGAACGGCTGGAGTGGAATCTGCAGTACTG
SSIV 18/10 PB2       TAACAGAAGATCCAGATGAGGGAACGGCTGGAGTGGAATCTGCAGTACTG
Haseluenne 2003      TAACAGAAGATCCGGATGAGGGAACGGCTGGAGTGGAATCTGCAGTACTG
Mexico 2009          TGACTGAAGATCCAGATGAAGGCACATCTGGGGTGGAGTCTGCTGTCCTG
                     * ** ******** ***** ** **  **** ***** ***** ** ***

SSIV 294/09 PB2      AGAGGGTTCCTAATTCTAGGGAAGGAAAATAAAAGATATGGACCAGCATT
SSIV 18/10 PB2       AGAGGGTTCCTAATTCTAGGGAAGGAAAATAAAAGATATGGACCAGCATT
Haseluenne 2003      AGAGGGTTCCTAATTCTAGGGAAGGAAAACAAAAGATATGGACCAGCATT
Mexico 2009          AGAGGATTTCTCATTTTGGGCAAAGAAGACAAGAGATATGGCCCAGCATT
                     ***** ** ** *** * ** ** *** * ** ******** ********

SSIV 294/09 PB2      GAGTATCAATGAACTGAGCAATCTTGCAAAAGGGGAGAAAGCCAACGTTT
SSIV 18/10 PB2       GAGTATCAATGAACTGAGCAATCTTGCAAAAGGGGAGAAAGCCAACGTTT
Haseluenne 2003      AAGTATCAATGAACTGAGTAATCTTGCAAAAGGGGAGAAGGCCAACGTTT
Mexico 2009          AAGCATCAATGAACTGAGCAATCTTGCAAAAGGAGAGAAGGCTAATGTGC
                     ** ************** ************** ***** ** ** **

SSIV 294/09 PB2      TGATAGGGCAAGGAGACGTGGTGTTGGTAATGAAACGGAAACGGGACTCT
SSIV 18/10 PB2       TGATAGGGCAAGGAGACGTGGTGTTGGTAATGAAACGGAAACGGGACTCT
Haseluenne 2003      TGATAGGGCAAGGGGACGTGGTGTTGGTAATGAAACGGAAACGGGACTCT
Mexico 2009          TAATTGGGCAAGGGGACGTAGTGTTGGTAATGAAACGAAAACGGGACTCT
                     * ** ******** ***** ***************** ************

SSIV 294/09 PB2      AGCATACTTACTGACAGCCAGACAGCGACCAAAAGAATTCGGTNGGCCAT
SSIV 18/10 PB2       AGCATACTTACTGACAGCCAGACAGCGACCAAAAGAATTCGGTTGGCCAT
Haseluenne 2003      AGCATACTTACTGACAGCCAGACAGCGACCAAAAGAATTCGGTTGGCCAT
Mexico 2009          AGCATACTTACTGACAGCCAGACAGCGACCAAAAGAATTCGGATGGCCAT
                     ****************************************  ******

SSIV 294/09 PB2      CAATTAGTGTTNAATTGTTTAAAAACGACCTTGTTTCTACT
SSIV 18/10 PB2       CAATTAGTGTTGAATTGTTTAAAAACGACCTTGTTTCTACT
Haseluenne 2003      CAATTAGTGTTGAATTGTTTAAAAACGACCTTGTTTCTACT
Mexico 2009          CAATTAGTGTCGAATTGTT----------------------
                     **********  *******
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Legend:
Nucleotide sequences of segment 2. Highlighted in red: position 44 in the translated amino acid sequence determines over privileged hu-hu transmission.

Appendix 8b

PB2 protein

```
SSIV 18/10 PB2       MERIKGLRDLMSQSRTREILTKTTVDHMAIIKKYTSGRQEKNPSLRMKWM
SSIV 294/09 PB2      MERIKELRDLMSQSRTREILTKTTVDHMAIIKKYTSGRQEKNPSLRMKWM
Haseluenne 2003      MERIKELRDLMSQSRTREILTKTTVDHMAIIKKYTSGRQEKNPALRMKWM
Mexico 2009          MERIKELRDLMSQSRTREILTKTTVDHMAIIKKYTSGRQEKNPALRMKWM
                     ***** ************************************:******

SSIV 18/10 PB2       MAMKYPITADRRIMEMIPERNEQGQILWSKTNDAGSDRVMVSPLAVTWWN
SSIV 294/09 PB2      MAMKYPITADRRIMEMIPERSEQGQILWSKTNDAGSDRVMVSPLAVTWWN
Haseluenne 2003      MAMKYPITADRRIMEMIPERNEQGQILWSKTNDAGSDRVMVSPLAVTWWN
Mexico 2009          MAMRYPITADKRIMDMIPERNEQGQTLWSKTNDAGSDRVMVSPLAVTWWN
                     ***:******:***:*****.**** ************************

SSIV 18/10 PB2       RNGPTADTVHYPKVYKTYFEKVERLKHGTFGPVHFRNQVKIRRRVDINPG
SSIV 294/09 PB2      RNGPTTDTVHYPKVYKTYFEKVERLKHGTFGPVHFRNQVKIRRRVDINPG
Haseluenne 2003      RNGPTTDTVHYPKVYKTYFEKVERLKHGTFGPVHFRNQVKIRRRVDINPG
Mexico 2009          RNGPTTSTVHYPKVYKTYFEKVERLKHGTFGPVHFRNQVKIRRRVDTNPG
                     *****:.**************************************** ***

SSIV 18/10 PB2       HADLNAKEAQDVIMEVVFPNEVGARILTSESQLTITKEKKEELQDCKIAP
SSIV 294/09 PB2      HADLNAKEAQDVIMEVVFPNEVGARILTSESQLTITKEKKEELQDCKIAP
Haseluenne 2003      HADLSAKDAQDVIMEVVFPNEVGARILTSESQLTITKEKKEELQDCKIAP
Mexico 2009          HADLSAKEAQDVIMEVVFPNEVGARILTSESQLAITKEKKEELQDCKIAP
                     ****.**:************************:****************

SSIV 18/10 PB2       LMVAYMLERELVRKTRFLPVAGGTSSVYIEVLHLTQGTCWEQMYTPGGEV
SSIV 294/09 PB2      LMVAYMLERELVRKTRFLPVAGGTSSVYIEVLHLTQGTCWEQMYTPGGEV
Haseluenne 2003      LMVAYMLERELVRKTRFLPVAGGTSSVYIEVLHLTQGTCWEQMYTPGGKV
Mexico 2009          LMVAYMLERELVRKTRFLPVAGGTGSVYIEVLHLTQGTCWEQMYTPGGEV
                     ************************.***********************:*
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SSIV 18/10 PB2      KNDDIDQSLIIAARNIVRRAIVSADPLASLLEMCHGTQIGGIRMVDILKQ
SSIV 294/09 PB2     KNDDIDQSLIIAARNIVRRAIVSADPLASLLEMCHGTQIGGIRMVDILKQ
Haseluenne 2003     KNDDVDQSLIIAARNIVRRAIVSADPLASLLEMCHSTQIGGIRMVDILKQ
Mexico 2009         RNDDVDQSLIIAARNIVRRAAVSADPLASLLEMCHSTQIGGVRMVDILRQ
                    :***:*************** ************.*****:******:*

SSIV 18/10 PB2      NPTEEQAVDICKAAMGLRISSSFSFGGLTFKRTSGSSVKKEEEVLTGNLQ
SSIV 294/09 PB2     NPTEEQAVDICKAAMGLRISSSFSFGGFTFKRTSGSSVKREEEVLTGNLQ
Haseluenne 2003     NPTEEQAVDICKAAMGLRISSSFSFGGFTFKRTSGSSVKREEEVLTGNLQ
Mexico 2009         NPTEEQAVDICKAAIGLRISSSFSFGGFTFKRTSGSSVKKEEEVLTGNLQ
                    **************:************:***********:**********

SSIV 18/10 PB2      ALKIRVHEGYEEFTMVGRRATAILRKATRRLIQLIVSGRDEQSIAEAIIV
SSIV 294/09 PB2     ALKIRVHEGYEEFTMVGRRATAILRKATRRLIQLIVSGRDEQSIAEAIIV
Haseluenne 2003     ALKIRVHEGYEEFTMVGKRATAILRKATRRLIQLIVSGRDEQSIAEAIIV
Mexico 2009         TLKIRVHEGYEEFTMVGRRATAILRKATRRLIQLIVSGRDEQSIAEAIIV
                    :****************:*********************************

SSIV 18/10 PB2      AMVFSQEDCMIKAVRGDLNFVNRANQRLNPMHQLLRHFQKDAKVLFQNWG
SSIV 294/09 PB2     AMVFSQEDCMIKAVRGDLNFVNRANQRLNPMHQLLRHFQKDAKVLFQNWG
Haseluenne 2003     AMVFSQEDCMIKAVRGDLNFVNRANQRLNPMHQLLRHFQKNAKVLFQNWG
Mexico 2009         AMVFSQEDCMIKAVRGDLNFVNRANQRLNPMHQLLRHFQKDAKVLFQNWG
                    ***************************************:*********

SSIV 18/10 PB2      IEPIDNIMGMIGILPDMTPSTEMSLRGVRISKTGVDEYSSTERVVVSIDR
SSIV 294/09 PB2     IEPIDNIMGMIGILPDMTPSTEMSLRGVRISKTGVDEYSSTERVVVSIDR
Haseluenne 2003     IEPIDNIMGMIGILPDMTPSTEMSLRGVRVSKTGVDEYSSTERVVVSIDR
Mexico 2009         IESIDNVMGMIGILPDMTPSTEMSLRGIRVSKMGVDEYSSTERVVVSIDR
                    **.***:********************:*:** *****************

SSIV 18/10 PB2      FLRVRDQRGNVLLSPEEVSETQGTEKLTITYSSSMMWEINGPESVLVNTY
SSIV 294/09 PB2     FLRVRDQRGNVLLSPEEVSETQGTEKLTITYSSSMMWEINGPESVLVNTY
Haseluenne 2003     FLRVRDQRGNVLLSPEEVSETQGTEKLTITYSSSMMWEINGPESVLINTY
Mexico 2009         FLRVRDQRGNVLLSPEEVSETQGTERLTITYSSSMMWEINGPESVLVNTY
                    ************************:********************:***

SSIV 18/10 PB2      QWIIRNWENVKIQWSQDPTMLYNKMEFEPFQSLIPKAARGQYSGFVRTLF
SSIV 294/09 PB2     QWIIRNWENVKIQWSQDPTMLYNKMEFEPFQSLIPKAARGQYSGFVRTLF
Haseluenne 2003     QWIIRNWETVKIQWSQDPTMLYNKMEFEPFQSLVPKAARGQYSGFVRTLF
Mexico 2009         QWIIRNWEIVKIQWSQDPTMLYNKMEFEPFQSLVPKATRSRYSGFVRTLF
                    ******** ************************:***:*.:*********

SSIV 18/10 PB2      QQMRDVLGTFDTAQIIKLLPFAAAPPEQSRMQFSSLTVNVRGSGLRILIR
SSIV 294/09 PB2     QQMRDVLGTFDTAQIIKLLPFAAAPPEQSRMQFSSLTVNVRGSGLRILVR
Haseluenne 2003     QQMRDVLGTFDTAQIIKLLPFAAAPPEQSRMQFSSLTVNVRGSGLRILIR
Mexico 2009         QQMRDVLGTFDTVQIIKLLPFAAAPPEQSRMQFSSLTVNVRGSGLRILVR
                    ************.***********************************:*

SSIV 18/10 PB2      GNSPVFNYNKTTKRLTVLGKDAGALTEDPDEGTAGVESAVLRGFLILGKE
SSIV 294/09 PB2     GNSPVFNYNKTTRRLTVLGKDAGALTEDPDEGTAGVESAVLRGFLILGKE
Haseluenne 2003     GNSPVFNYNKATKRLTVLGKDAGALTEDPDEGTAGVESAVLRGFLILGKE
Mexico 2009         GNSPVFNYNKATKRLTVLGKDAGALTEDPDEGTSGVESAVLRGFLILGKE
                    **********:*:********************:****************
SSIV 18/10 PB2      NKRYGPALSINELSNLAKGEKANVLIGQGDVVLVMKRKRDSSILTDSQTA
SSIV 294/09 PB2     NKRYGPALSINELSNLAKGEKANVLIGQGDVVLVMKRKRDSSILTDSQTA
Haseluenne 2003     NKRYGPALSINELSNLAKGEKANVLIGQGDVVLVMKRKRDSSILTDSQTA
Mexico 2009         DKRYGPALSINELSNLAKGEKANVLIGQGDVVLVMKRKRDSSILTDSQTA
                    :************************************************

SSIV 18/10 PB2      TKRIRLAIN
SSIV 294/09 PB2     TKRIRXAIN
Haseluenne 2003     TKRIRLAIN
Mexico 2009         TKRIRMAIN
                    ***** ***
```

Legend:
Amino acid sequences of PB2 protein. 44S (highlighted in red) is an indicator for privileged hu-hu transmission.

Appendix 9

List of manual sequence corrections

Nonstructural Protein
SIV 132/09: Position 83: insertion of A
Position 880: replacement of N with T
SIV247/09: Position 167: insertion of C
Position 168/169: replacement of NN with CC
SIV 270/09: Position 10: replacement of M with C
Position 11: insertion of A
Position 461: replacement of G with A
Position 517: replacement of T with C
Position 901: replacement of G with K
SIV 54/10: Position 125: insertion of G
Position 127/128: replacement of NN with GG

Matrix Protein
SIV 132/09: Position 10: insertion of C
Position 1037: replacement of N with T
Position 1038: insertion of G
SIV 206/09: Position 359: replacement of A with M
Positon 383: replacement of G with R
Position 484: replacement of T with Y
Position 620: replacement of A with R
Position 714: replacement of A with R
Position 761: replacement of G with R
Position 766: replacement of C with Y
Position 1037: replacement of N with T
SIV 226/09: Position 1038: replacement of N with T
SIV 294/09: Position 1032: deletion of N
SIV 45/10: Position 1034: replacement of N with C
Position 1036: replacement of N with A
SIV 54/10: Position 1035: replacement of N with T

Neuraminidase
SIV 206/09: Position 547: replacement of N with A
Position 548: insertion of G
Position 1468: replacement of N with T
Position 1469: insertion of G
SIV 270/09: Position 1468: insertion of T
Position 1478: insertion of T
SIV 294/09: Position 80: replacement of A with G
Position 205: replacement of T with A
Position 274: replacement of t with C
Position 1468: replacement of T with N
Position 2478: insertion of T
SIV 54/10: Position 1464: replacement of N with C
Position 1465: replacement of N with T
Position 1468: replacement of N with T

Nucleoprotein
SIV 246/09: Position 825-826: replacement of TN with NT
SIV 206/09: Position 1128: replacement of C with T
Position 1329: replacement of C with T
Position 1576: replacement of N with T
SIV 246/09: Position 760: replacement of NA with AM
Position 1278: replacement of S with W

Haemagglutinin
SIV 132/09: Position 519: insertion of C
SIV 206/09: Position 519: insertion of C
SIV 270/09: Position 1787: replacement of N with T
Position 1788: insertion of G
SIV 45/10: Position 730: replacement of G with K

PB1
SIV 294/09: Position 744: insertion of A

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